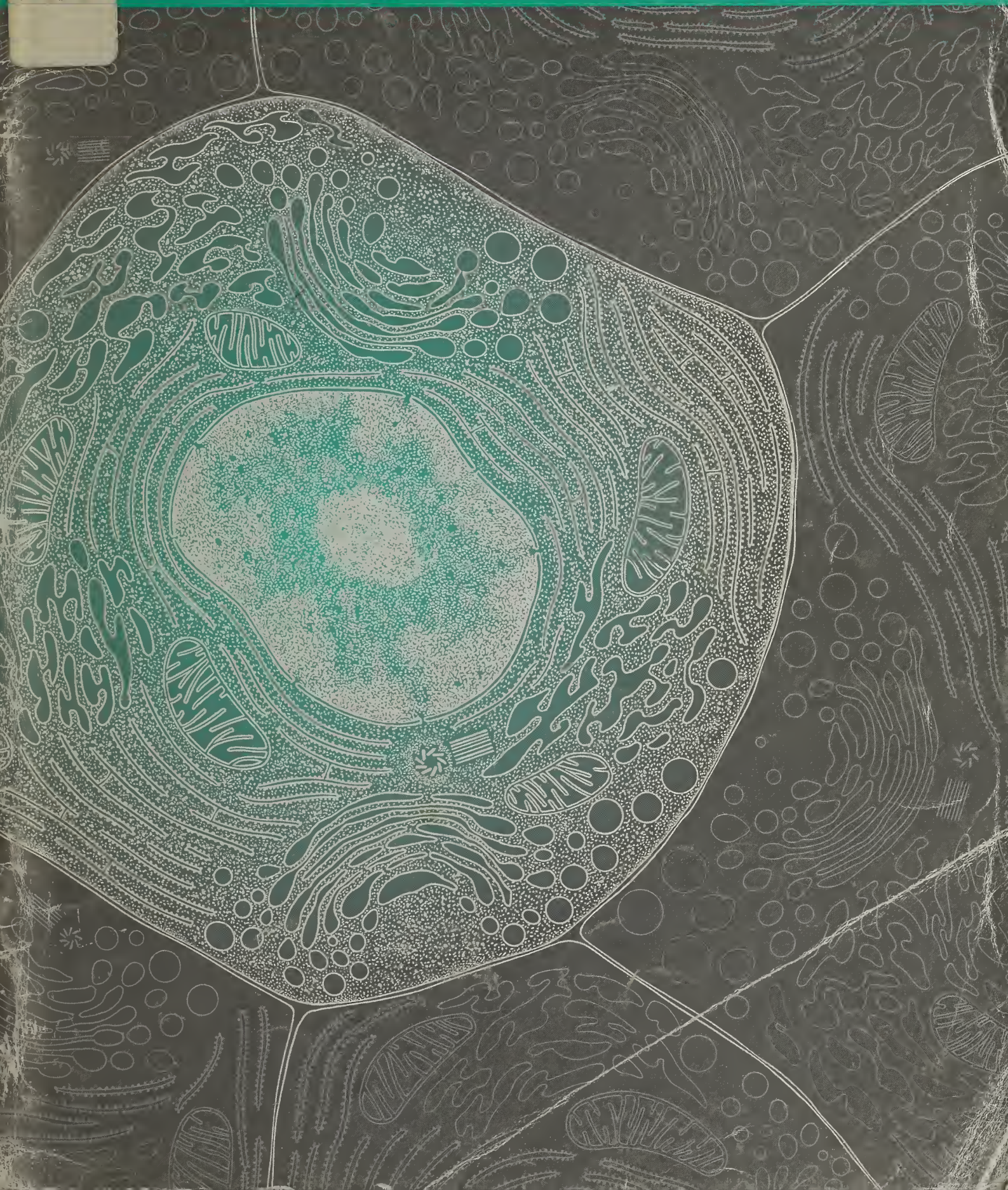
INSIDE THE CELL

NATIONAL INSTITUTES OF HEALTH National Institute of General Medical Sciences



What Is NIGMS ?

The National Institute of General Medical Sciences (NIGMS) is unique among the components of the National Institutes of Health (NIH) in that its main mission is the advancement of the basic biomedical sciences. It supports selected research and research training programs in areas that underlie all medical investigation, such as cellular and molecular biology and genetics. Knowledge resulting from this work contributes directly to the progress of research on specific diseases in the other components of NIH. NIGMS also develops and supports interdisciplinary studies in biophysics, pharmacology, biological chemistry, physiology, and developmental biology. Many of the researchers mentioned in this brochure worked with NIGMS support.

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THE FUNDAMENTAL UNIT OF LIFE

The cell is the fundamental unit of life. Your health depends on what happens within the many different types of cells that make up your body. The health of your cells depends, in turn, on the function of millions of critical molecules.

Since the mid-1940's, biomedical researchers have made enormous progress in identifying and understanding these molecules and how they interact in many cellular processes. Much of this research was "basic"— aimed simply at learning how living systems work. The fundamental knowledge developed through this research can lead to new ways to diagnose, treat, cure, or prevent disease.

A stunning example of how basic cell biology research is moving toward practical application is found in studies of the cycle of cell growth and division. In recent years, many details of the biochemical mechanisms involved in the normal cell cycle have been discovered. Scientists have found the cell cycle to be regulated by highly complex interactions between pairs of proteins that belong to two general families. Work is proceeding at top speed to determine all of the many molecular interactions and the order in which they occur during the cell cycle. This work is yielding an understanding of the normal processes of growth and development that will, in turn, aid researchers seeking to treat diseases in which these processes go awry.

Other scientists are discovering direct connections between cell cycle regulation and cancer. This research is beginning to demonstrate the specific role of oncogenes, genes that are directly involved in the development of cancer, and tumor suppressor genes, which are involved in cancer when their normal inhibitory functions are disrupted.

To help readers understand some of the exciting biomedical research being conducted today, *Inside the Cell* provides an overview of the basic facts of cell biology. The brochure also contains some history of key scientific discoveries.

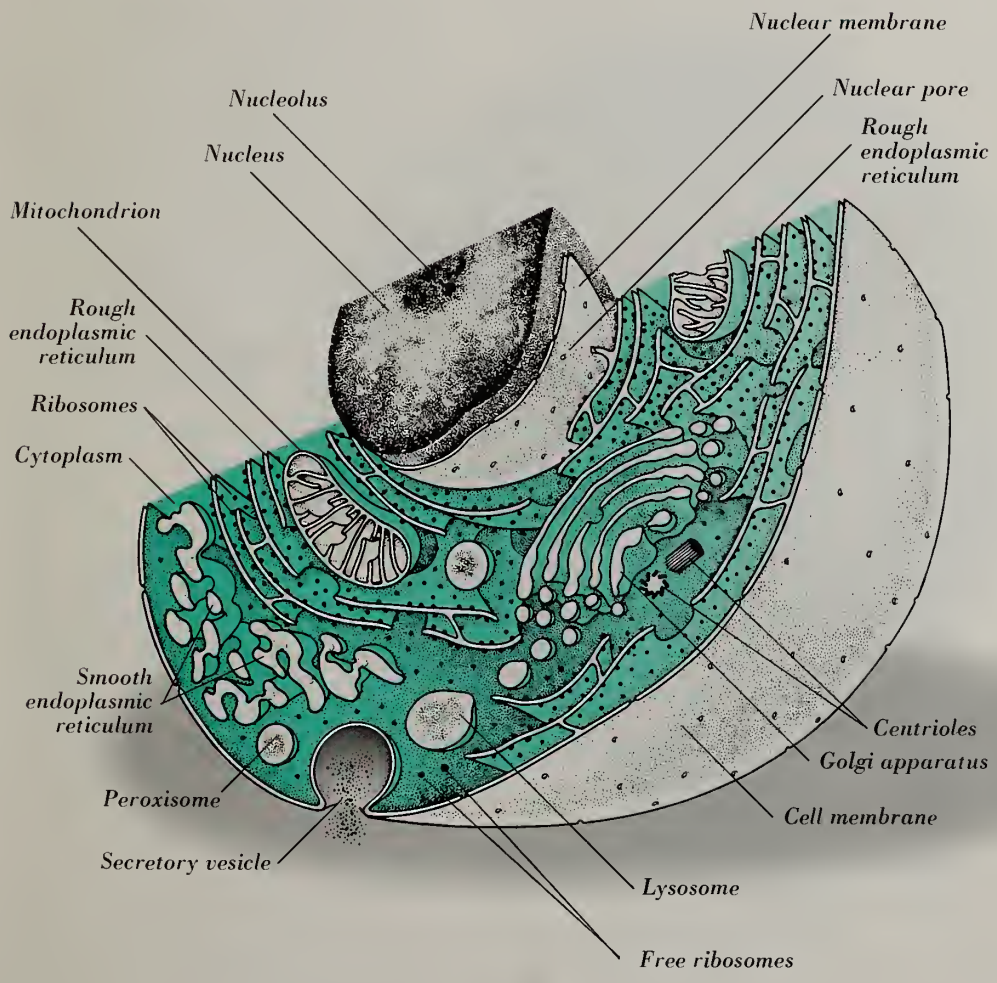
Many scientists agree that the history of modern cell biology began with a convergence of improved techniques in microscopy and biochemistry. In the 1950's, as scientists working in these fields began to collaborate, they started to develop our current picture of the cell as a complex and highly organized entity.

They found that a typical cell is like a miniature body containing tiny "organs," called organelles. One organelle is the command center, others provide the cell with energy, while still others manufacture proteins and additional molecules that the cell needs to survive and to communicate with the world around it. The entire cell is enclosed in a fine

"skin," its surface membrane. This membrane not only keeps the cell intact, it also provides channels that open and close to allow selected molecules into and out of the cell.

Scientists are seeking to learn more about the ways cells respond to outside signals, which are often conveyed when molecules bind to special receptors in cell membranes. Because the shape of a molecule plays a large part in determining its function, scientists are also keenly interested in determining the shape of important molecules and the rules that cause a string of chemicals to fold into a specific molecular shape.

To understand cellular function, most scientists study parts of specific biochemical pathways, such as the cell cycle, that involve individual molecules, cells, groups of cells, and whole organisms. The goal is, of course, to be able to put all the parts together to understand normal cellular activities and how they malfunction in disease. ■



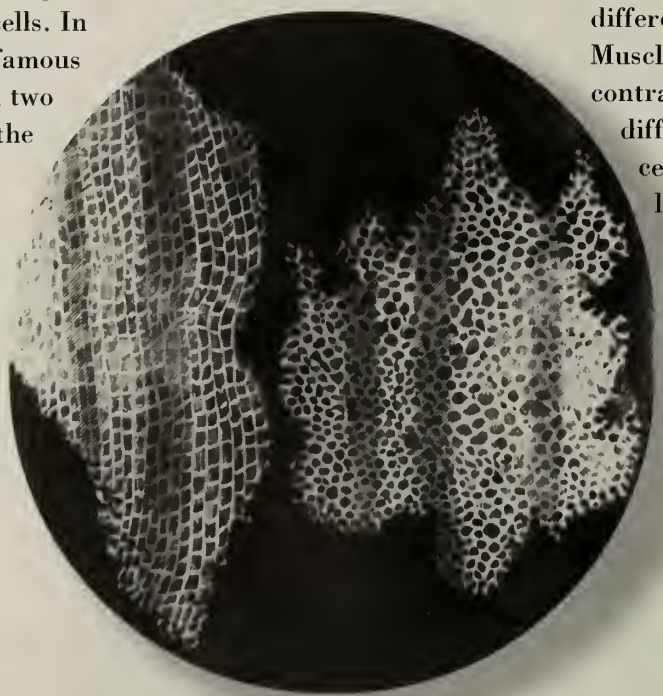
This drawing of an idealized animal cell is based on photographs taken with powerful electron microscopes. Within the cell's membrane are such organelles as the mitochondria (energy producers), the rough endoplasmic reticulum (a site of protein production), the Golgi apparatus (a protein sorter), and the largest organelle, the nucleus (which contains the hereditary material DNA). In addition to these organelles, cells also contain an elaborate network of protein filaments called the cytoskeleton (not shown here) that anchor the organelles, maintain the cell's shape, and direct intracellular traffic.

In 1665, the English physicist Robert Hooke looked at a sliver of cork through a microscope lens and noticed some "pores" or "cells" in it. Hooke believed the cells had served as containers for the "noble juices" or "fibrous threads" of the once-living cork tree. He thought these cells existed only in plants, since he and his scientific contemporaries had observed the structures only in plant material.

Nearly two centuries later, scientists began to develop the idea that every living thing is made up of cells. In 1838, during a now-famous dinner conversation, two German scientists—the botanist Matthias Schleiden, who had been studying plant cells, and the zoologist Theodor Schwann, who had been examining the nervous tissue of animals—realized that the similarities between the structures they had been investigating were too strong to be accidental. In 1847, Schwann wrote a paper describing how all animal tissue, including bone, blood, skin, muscle, and glands, is composed of cells. Even sperm and eggs are cells. Schleiden elaborated on this idea as it applied to plants. A German pathologist, Rudolph Virchow, is given credit for being the first to state, in 1858, what became known as the cell theory: "Every animal appears as a sum of vital units, each of which bears in itself the complete characteristics of life."

The cell theory united plant and animal sciences by recognizing that the cell is the fundamental component of all living organisms, from orchids and earthworms to human beings. It provided an intellectual framework that revealed the hidden similarities of form and function in extremely diverse organisms, and it gave scientists a way of making sense out of the bewildering array of living creatures. But what is a cell?

Obviously, there are major differences among cell types. Muscle cells, which can contract, have to be quite different from liver or bone cells. Nerve cells have long, thin fibers that, in humans, may extend more than 3 feet from the spinal cord to the toes, while blood cells have no projecting



This drawing of cork tissue, as seen under a simple microscope, appeared in Robert Hooke's 1667 book, Micrographia. Hooke named the compartments "cells."

fibers at all. Plant cells have a unique ability to use light as a source of energy.

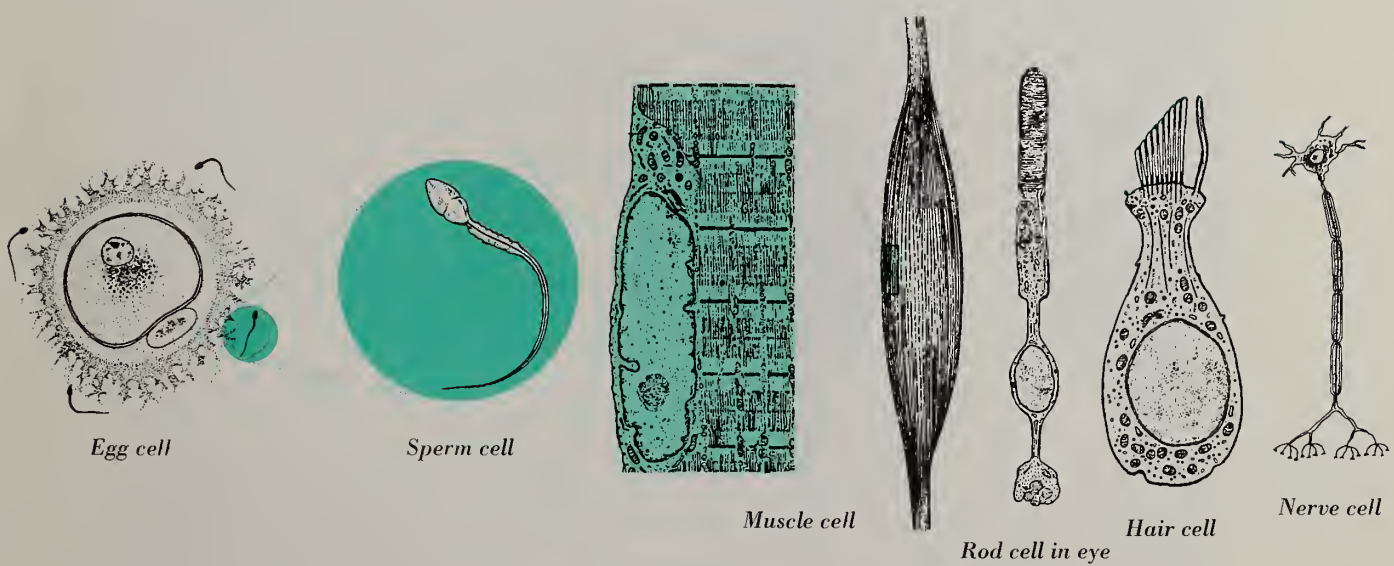
Then what do all these cells have in common? Discovering their shared properties was difficult. At first, scientists thought that the cell was just a blob of jelly, or some primordial soup enclosed in a bag. They named the jelly "protoplasm." For a long time they could not find anything in the protoplasm, which is now known as the "cytoplasm."

Part of the difficulty in studying cells, of course, is due to their extremely small size. The cells of multicellular organisms are impossible to see with the unaided eye. Schleiden and Schwann, like cell biologists before and after, relied upon microscopes to enlarge the image of cells so that they could be studied. Microscopes employ one or more curved lenses and a source of illumination (typically white light) to magnify cells.

One of the most remarkable early microscopists was a Dutch draper named Anton van Leeuwenhoek, who ground his own lenses as a hobby. Van Leeuwenhoek, who once made a lens from a grain of sand, used simple (single-lensed) microscopes to examine everything from pond water to the scum on his teeth.

In 1702, van Leeuwenhoek reported to the Royal (Scientific) Society of London that he had observed "a little clear sort of light in the middle" of a fish blood cell

The variety of human cells. After fertilization by a sperm, a single human egg cell divides again and again into many kinds of specialized cells whose structures vary according to the functions they fill. Some nerve cells, for example, are 3 feet long to reach from spine to toe. The orderly structure of typical skeletal muscle is shown here in such detail that if the entire muscle cell were drawn to the same scale as the fragment shown, it might be 1,000 feet long.



Egg cell

Sperm cell

Muscle cell

Rod cell in eye

Hair cell

Nerve cell

Anton van Leeuwenhoek (1632-1723) was an early microscopist who ground his own lenses as a hobby and was the first to observe such living cells as sperm and pond water microorganisms.

he had been examining. This description of what was later called the cell's nucleus was the first suggestion that animal cells had an internal structure. Throughout the 18th and 19th centuries, improvements in microscopes and techniques for selectively staining cell parts enabled cell biologists to distinguish other particles within the cell. However, researchers could not study these minute flecks in detail because they met an insurmountable obstacle: the wavelength of light.

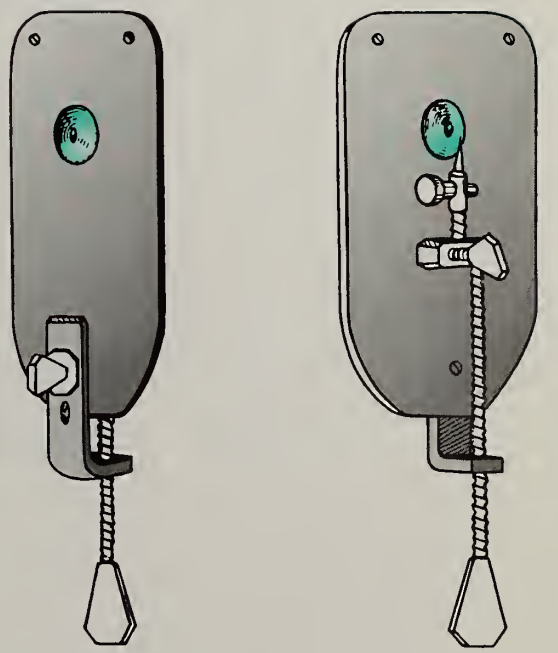
A light microscope—even one with perfect lenses and perfect illumination—simply cannot be used to distinguish objects that are smaller than half the wavelength of light. White light has an average wavelength of 0.55 micrometers, half of which is 0.275 micrometers. (One micrometer is a thousandth of a millimeter, and there are about 25,000 micrometers to an inch. Micrometers are also called microns.) Any two lines that are closer together than 0.275 micrometers will be seen as a single line, and any object with a diameter smaller than 0.275 micrometers will be invisible—or, at best, show up as a blur.

Although the nucleus of a typical human cell is relatively large (about 7 micrometers in diameter), most organelles vary from a width of only 1 micrometer to structures so fine that they must be measured in nanometers (which are 1,000 times smaller than micrometers), or even in angstrom units (10 times smaller than nanometers). To see such tiny particles under a microscope, scientists must bypass light altogether and use a different sort of "illumination," one with a shorter wavelength.

The invention of the electron microscope in the 1930's filled the bill. In this kind of microscope, electrons are speeded up in a vacuum until their wavelength is extremely short—only one hundred-thousandth that of white light. Beams of these fast-moving electrons are focused on a cell sample and are absorbed or scattered by the cell's parts so as to form an image on an electron-sensitive photographic plate.

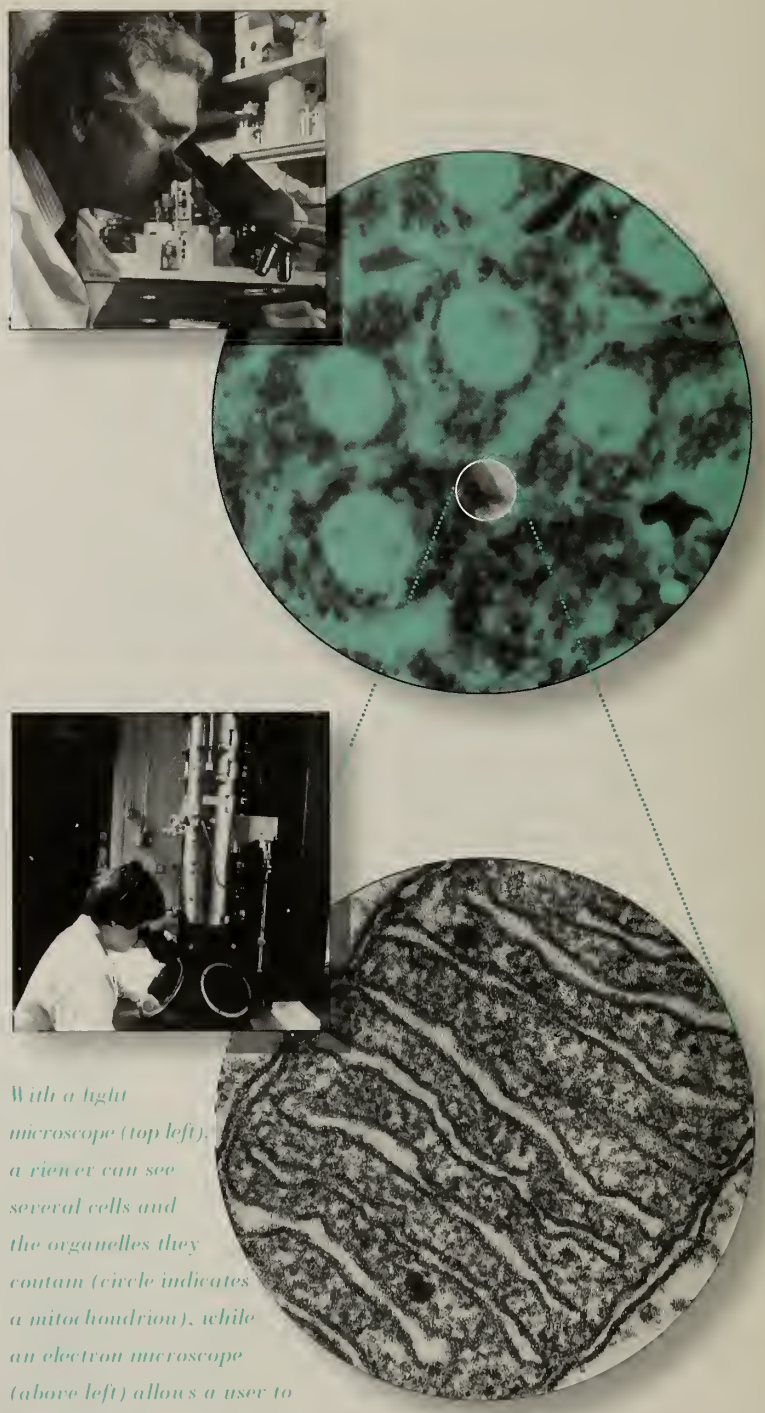
If pushed to the limit, electron microscopes can make it possible to view objects as small as the diameter of an atom. Most electron microscopes used to study biological material can "see" down to about 10 angstroms—an incredible feat, for although this does not make atoms visible, it does allow researchers to distinguish individual molecules of biological importance. In effect, it can magnify objects up to 1 million times. Nevertheless, all electron microscopes suffer from a serious drawback. Since no living specimen can

This is the actual size of a typical microscope built by van Leeuwenhoek. He peered through the tiny lens opening on one side of a metal plate (left) to see the specimen mounted on the point of a pin on the other side (right). The specimen could be moved into focus by a system of screws.

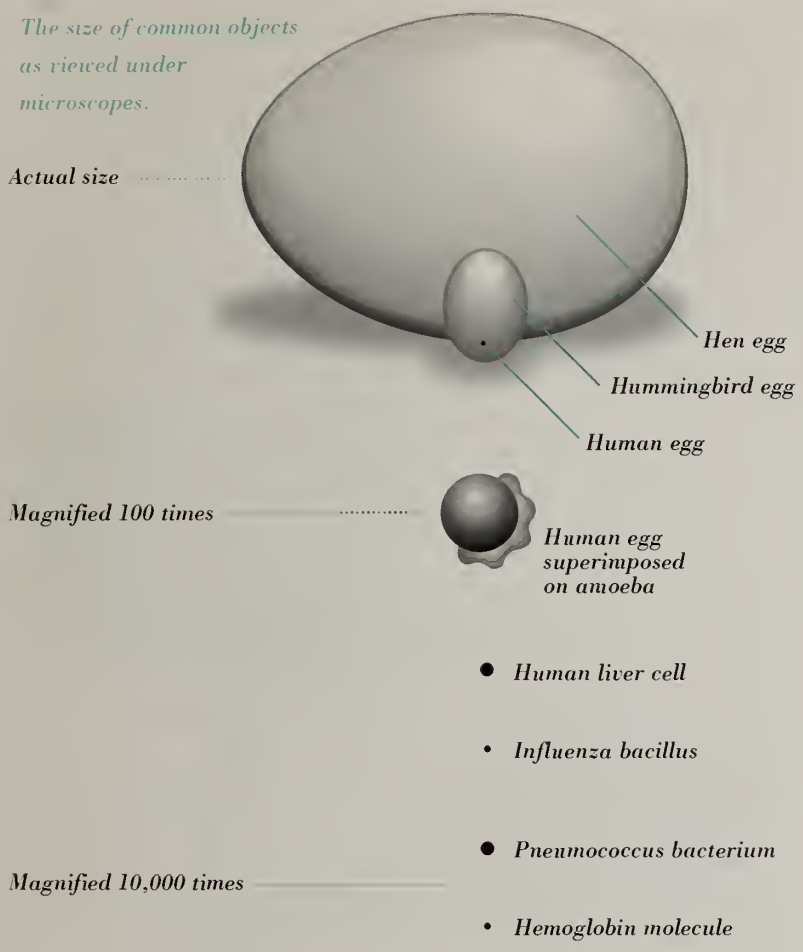


survive under their high vacuum, they cannot show the ever-changing movements that characterize a living cell.

The first electron microscopes were used to study crystals and were impractical for the study of cell structure. Cell researchers had to learn how to cut extremely thin slices of cells, sometimes down to a thickness of only a few hundred angstroms, so that electrons could pass through them. Also, to ensure contrast between different parts of the otherwise transparent cell, new staining techniques had to be devised. These techniques use special metal-containing



With a light microscope (top left), a viewer can see several cells and the organelles they contain (circle indicates a mitochondrion), while an electron microscope (above left) allows a user to see objects of interest (in this case a mitochondrion) in much greater detail.



The size of common objects as viewed under microscopes.

compounds that are absorbed to differing extents by various cell parts. The cell sections also had to be "fixed" in new ways, to preserve them, and had to be embedded in new kinds of materials (mostly transparent plastic). Altogether, it was not until the early 1950's that electron microscopes began to be used routinely for cell biological studies. While microscopists were looking through their instruments at smaller and smaller particles in cells and attempting to understand their structure, another group of scientists was pursuing an entirely different, but equally important, line of research—biochemistry. ■

Units of size commonly used in cell biology.

Unit	Equal to	Used to Measure
Centimeter	1/100 meter	*Objects visible to the eye*
Millimeter	1/10 centimeter	*Very large cells*
Micrometer (micron)	1/1000 millimeter	*Most cells, large organelles*
Nanometer	1/1000 micrometer	*Small organelles, large molecules*
Angstrom	1/10 nanometer	*Molecules, atoms*

Modern Light Microscopy Gives a Clear View of Cell Structure and Movement

Using a microscope the size of his palm, Anton van Leeuwenhoek was able to study the movements of one-celled organisms. Modern descendants of van Leeuwenhoek's light microscope can be over 6 feet tall, but they continue to be indispensable to cell biologists because, unlike electron microscopes, light microscopes enable the user to see living cells in action. The primary challenge for light microscopists since van Leeuwenhoek's time has been to enhance the contrast between pale cells and their paler surroundings so that cell structures and movement can be seen more easily. To do this they have devised ingenious strategies involving video cameras, polarized light, digitizing computers, and other techniques that are yielding vast improvements in contrast, fueling a renaissance in light microscopy.

A polarizer causes light waves to move in parallel planes, thus reducing the distortion that results when light scatters across a magnified object. This technique was first used in the 1950's, and it provided many new clues to cellular activities, particularly the intricacies of cell division. Further enhancements in visualizing the cell's interior came in the early 1980's, when microscopists Shinya Inoué of the Marine Biological Laboratory in Woods Hole, Massachusetts,

and the late Robert Allen of Dartmouth College began using video cameras to study living cells. Unlike the eye, a video camera can enhance an image to "see" objects clearly even when the contrast between subject and background is very poor. Inoué and Allen used video cameras to watch food-containing sacs and other structures in the cell move rapidly along slender, track-like organelles. Video images can now be further enhanced by digitizing computers, which, when attached to a video camera, scan the cell, break down the image into light and dark bits, and then reconstruct the image so that "visual noise" (grayness) is subtracted, while objects of interest are highlighted.

Another type of microscope, called the confocal microscope, is having a great impact on the study of cell structure. A confocal microscope passes a beam of light over a tiny portion of a cell, then focuses the light that reflects off the specimen through a pinhole. A sharply focused, three-dimensional image of a cell or cell structure can be built up by recording the intensity of the light beam coming off each scanned point and then reconstructing the whole image on a viewing screen. Because confocal microscopes can be used on living cells, they allow researchers to watch cell movements and the interactions of neighboring cells. ■

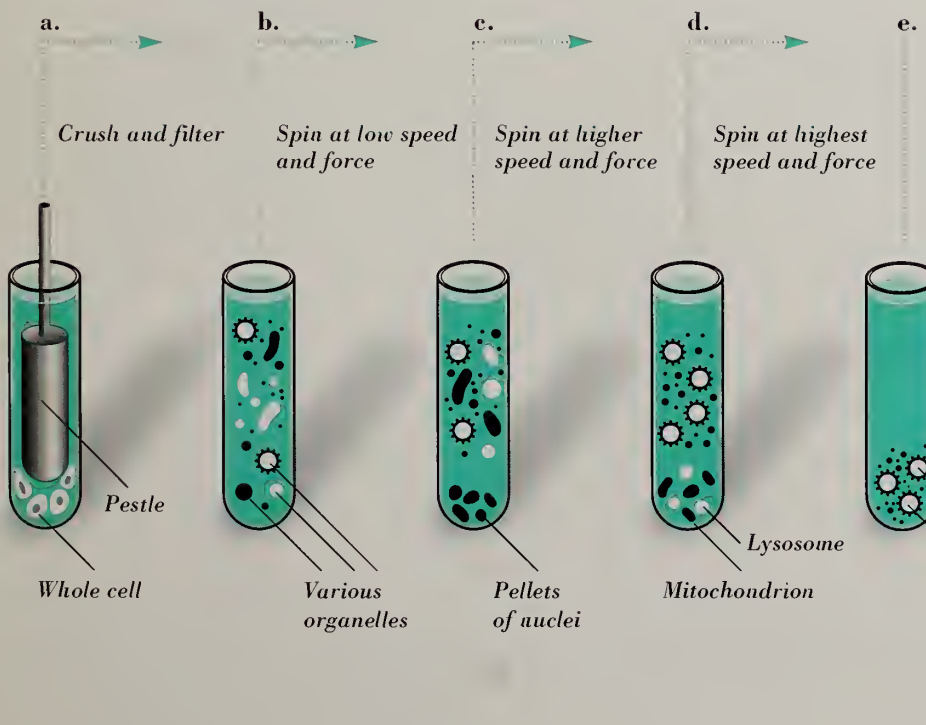
The study of biochemistry goes back to Antoine Lavoisier, the 18th-century French scientist who explained the role of oxygen in the metabolism of food to provide energy in both plants and animals, established the composition of water and other compounds, and introduced methods of measuring aspects of chemical reactions, thereby laying the foundation for modern chemistry.

In the 19th century, biochemists isolated and identified many cellular chemicals—for example, hemoglobin, the red pigment in blood, and chlorophyll, the green pigment in plants. They discovered that compounds taken from animal tissue consisted of many of the same chemical elements as nonliving materials. They isolated the nucleic acids DNA and RNA, which are now known to govern heredity and protein synthesis. They began to study proteins, especially enzymes, which catalyze chemical reactions in cells.

When dealing with cells, the biochemists behaved quite unlike the microscopists, who had enormous respect for the details of the cell's structure. The biochemists simply ground up, or homogenized, large quantities of cells to release their contents into a solution and then analyzed the mixture (called the homogenate). Often, the homogenate was fractionated, or separated, into individual components. Usually this was done with a centrifuge, a machine that separates particles according to their size and density by whirling them around at high speeds. The largest and heaviest particles move to the bottom of the container most rapidly, followed by somewhat smaller and lighter components, until after a time there remain only the smallest and lightest particles at the top.

In 1925, a Swede, Theodor Svedberg, developed an instrument that would prove at least as revolutionary as the electron microscope: the ultracentrifuge, a machine that could spin its samples at such high speeds and with such force (it could attain hundreds of thousands of times the force of gravity) that many of the smaller and lighter components of the cell and even proteins and nucleic acids could be collected separately and studied for the first time.

The significance of this new instrument did not become apparent until years later. For several decades, quite a few biochemists studied the chemical reactions of the cell without realizing that the microscopists could actually see many of the particles they were analyzing. Finally, in the 1950's, the two groups began to edge closer together. By that time, electron-microscopic techniques had been refined. As the microscopists and biochemists began to communicate, there was an avalanche of discoveries about the world within the cells of animals and plants. A whole new vocabulary had to be developed for the cellular structures that the electron microscope and ultracentrifuge revealed. Cell biologists began to define many of the general characteristics that cells share, and to discern the mechanisms that cells use to make proteins and other vital molecules. Understanding these mechanisms, in turn, enabled them to begin to identify specific steps in critical biochemical pathways. Some of these pathways will be mentioned in the following sections on specific cellular organelles. ■



How a centrifuge is used to isolate cell components. To separate the various particles in cells, biochemists begin by placing whole cells in a solution and then breaking the cells with a pestle or with high-frequency sound waves (a).

The mixture is then filtered to remove unbroken cells. At this point, the cell organelles and fragments are free-floating (b).

As the sample is spun in the centrifuge at increasingly higher speeds and force, the organelles begin to settle to the bottom depending on their size and density. First to settle out are pellets of nuclei (c).

At forces greater than 10,000 times that of gravity, pellets of mitochondria and lysosomes sink to the bottom (d).

Finally, at very high speeds and at forces 100,000 times that of gravity, the very lightest particles and organelles begin to settle out (e).

Obtaining Molecules for Study

To a large extent, the progress made since the 1950's in all areas of biology has been dependent on the development of techniques to obtain, grow, and purify sufficient quantities of specific types of cells and molecules, as well as to separate cellular components in centrifuges. In the late 1960's, scientists devised reliable methods of growing cells in the laboratory. Eventually, they also learned how to grow cells in various chemically defined solutions. These advances allowed researchers to study and compare biochemical processes in different types of cells and to determine the molecular details of many complex cellular activities.

Major improvements have also been made in sorting proteins and nucleic acids. A technique called column chromatography separates fragments of nucleic acids or proteins according to their size, electrical charge, and other important characteristics. This process uses a hollow column that is filled with a material through which the molecules in a solution move at different speeds. This allows a researcher to

collect the molecules separately as they pass out of the column. If necessary, additional methods of purifying the protein can then be used and its biological activity can be examined in detail.

Another technique, called gel electrophoresis, uses electrical currents to cause protein or nucleic acid molecules to travel through a semi-solid gel according to their size and charge. The separated molecules can then be transferred onto a sheet of special paper, where various methods can be used to detect each molecule or fragment. Electrophoresis is especially useful for the analysis and comparison of samples containing many different nucleic acid fragments or proteins.

In addition to the above techniques, cell biology has been revolutionized by recombinant DNA technology. Often called genetic engineering, recombinant DNA technology enables scientists to grow large quantities of cells that have been genetically altered to make a particular protein (usually a protein normally found in another organism) and then to harvest the protein from the cells. Often, bacteria and yeast cells are used to produce these proteins.

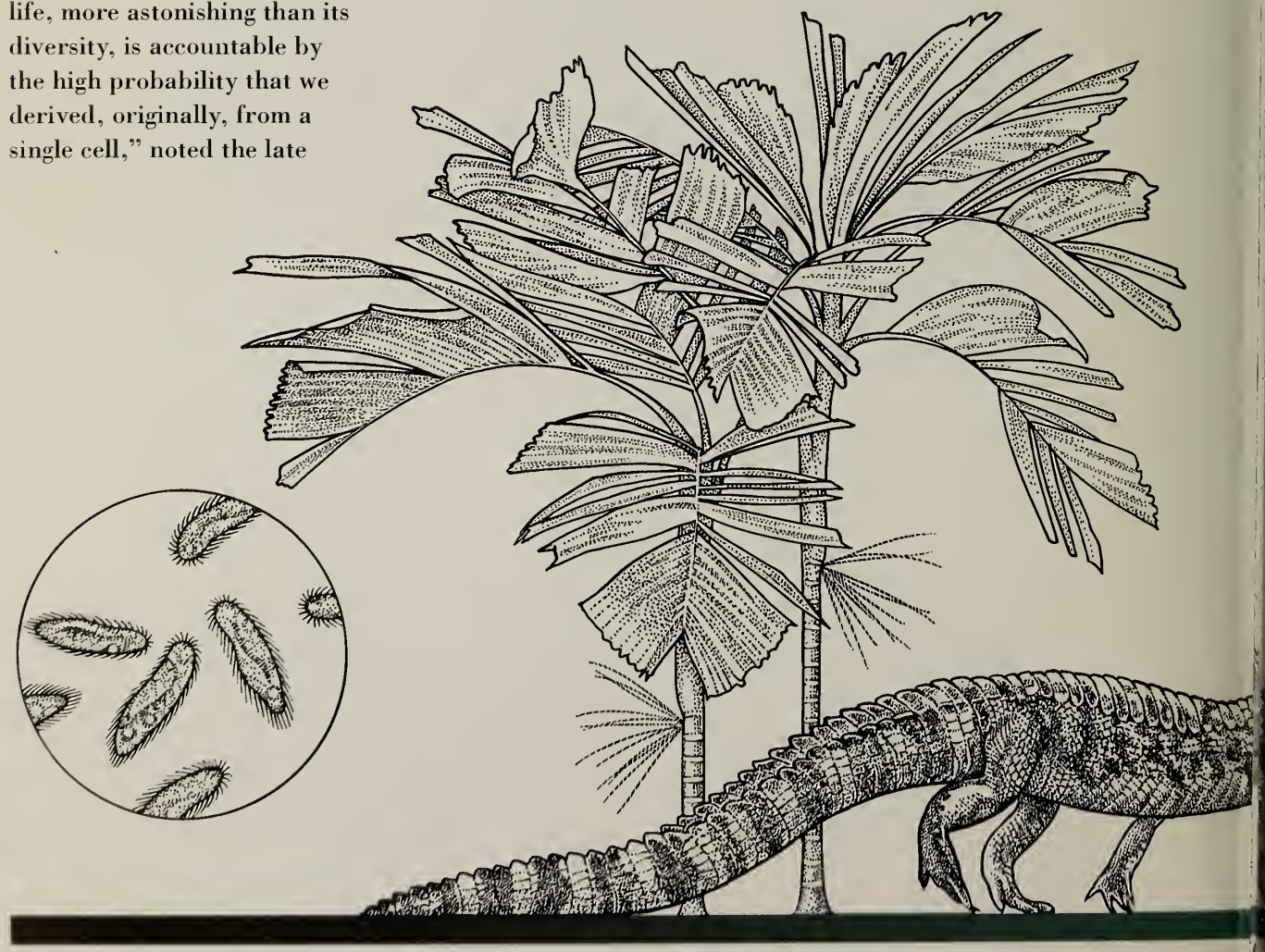
Through the use of recombinant DNA technology, scientists have discovered many new classes of genes and proteins and are able to compare the genetic material of species as different as humans and bacteria. This helps them determine the functions of proteins and regions within proteins and to piece together their roles in complex systems. Recombinant DNA technology has been augmented by another technique, called PCR (polymerase chain reaction), which allows researchers to make many copies of DNA segments without first having to grow these segments in bacteria or other organisms. ■

All cells—whether from a bacterium, plant, mouse, or human—are made of the same basic materials: nucleic acids, proteins, carbohydrates, water, fats, and salts. For this reason, scientists seeking to understand both normal and disease processes in humans can learn a great deal by studying similar systems in "model organisms" like bacteria, slime molds, yeast, and fruit flies.

"The uniformity of the earth's life, more astonishing than its diversity, is accountable by the high probability that we derived, originally, from a single cell," noted the late physician and author Lewis Thomas in *The Lives of a Cell*. "It is from the progeny of this parent cell that we take our looks; we still share genes around, and the resemblance of the enzymes of grasses to those of whales is a family resemblance."

The genetic material in all living cells is deoxyribonucleic acid (DNA), a large molecule that directs the making of duplicate cells. DNA also directs the building of proteins according to a code. Even the simplest living cells—the mycoplasma—contain a relatively large amount of DNA, enough to code for perhaps 1,000 different proteins. Every human cell has about 6 feet of very tightly wound DNA strands contained within its nucleus, and every adult carries billions of miles of ultrathin DNA strands in his or her body.

Each cell is separated from the rest of the world by a membrane so thin that it

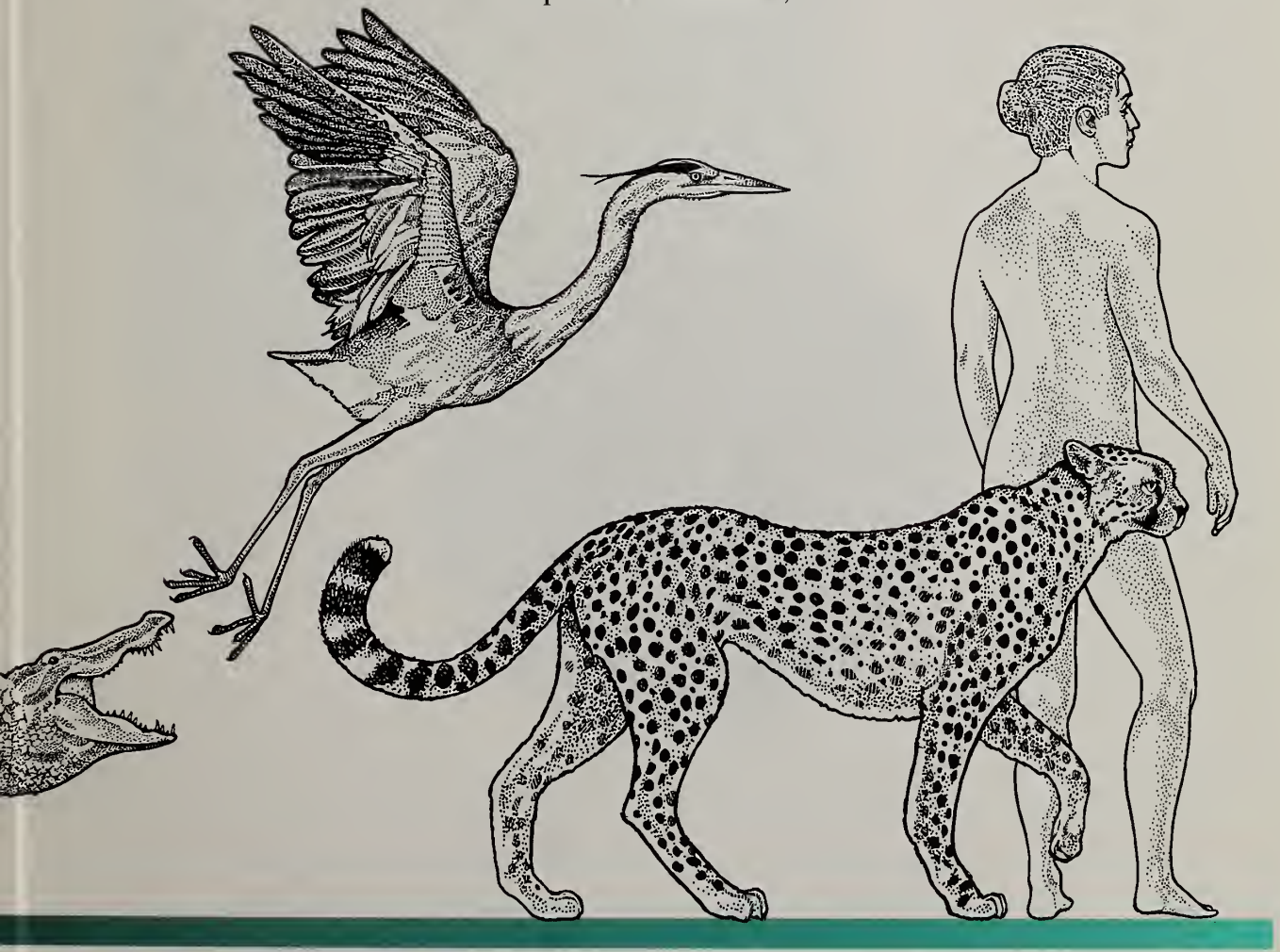


cannot be seen under a light microscope. Despite its thinness, the surface membrane is exceedingly sturdy, controlling everything that goes into and out of the cell and relaying vital messages. Similar membranes enclose or make up a large number of the cell's organelles.

There is a fundamental distinction between two of the major categories of cells. Prokaryotic cells, which include bacteria, mycoplasma, and cyanobacteria (blue-green algae), do not have a membrane around their nuclear region. Eukaryotic cells, in contrast, have two membranes separating the nucleus from the cytoplasm, as well as many other internal membranes to segregate their organelles. The cells of all animals and plants are eukaryotic.

Only eukaryotic cells are able to form large, multicellular systems—an important step up the evolutionary ladder. And while, in general, prokaryotic organisms produce only exact duplicates of themselves, higher eukaryotic organisms are capable of differentiation into many kinds of cells. This gives eukaryotes certain obvious advantages. However, prokaryotes have advantages of their own: simpler nutritional requirements, resistance to adverse conditions, and much more rapid growth and division.

The main job of most cells is to manufacture proteins. In eukaryotes, protein production begins in the most prominent organelle—the nucleus. ■

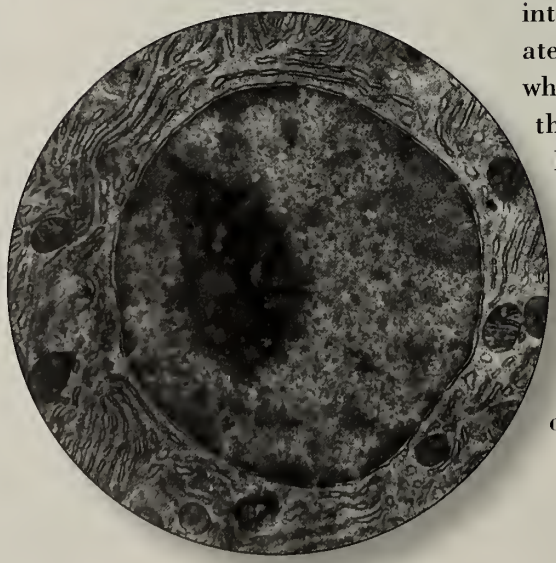


The nucleus is the biggest, densest, and most obvious structure in the eukaryotic cell—the first to be recognized by microscopists and the first to be isolated in the biochemists' centrifuge.

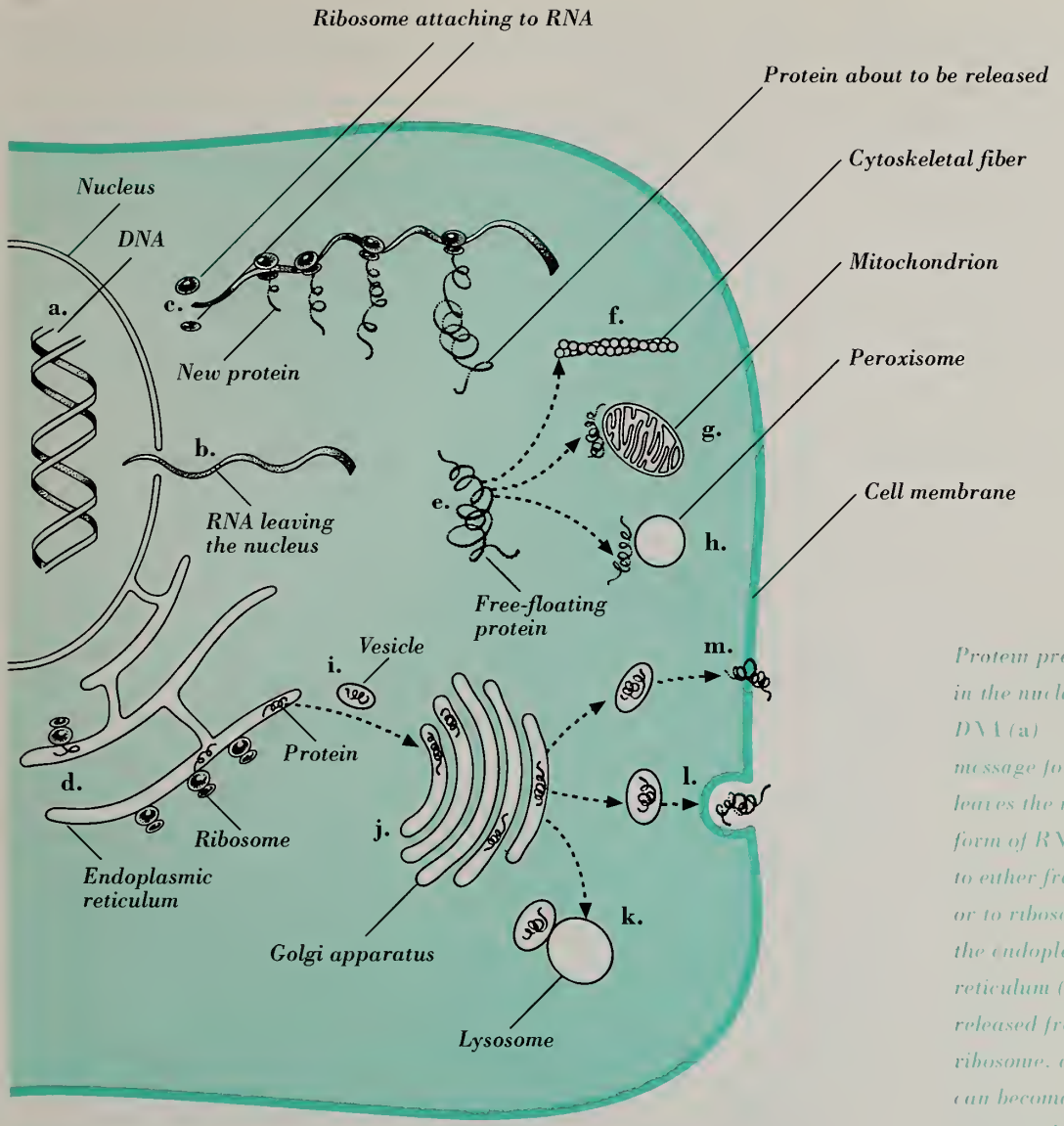
For many years, nobody knew what the nucleus did. In the 19th century, several researchers noted that before a cell divided, the nucleus divided. But it was not until the beginning of the 20th century that scientists grasped the connection between the rodlike chromosomes (tightly packed bundles of DNA and proteins) that had been observed in the nucleus and the transmission of hereditary traits. At that point, the importance of the nucleus became clear.

The nucleus is the cell's command center. The chromosomes contain the genes (made of DNA) that give directions for everything the cell is and will be, and thus control the cell's reproduction and heredity. DNA is a deceptively simple molecule, consisting of a sequence of subunits called bases. The bases are linked together to form a double helix that can be visualized as an immensely long, corkscrew-shaped ladder. Each rung in the ladder is made up of two bases joined together by chemical bonds, and the ends of the rung are attached to chains of chemically bonded sugar and phosphate molecules that are like the upright rails of the ladder. A unit of DNA containing one sugar molecule, one phosphate molecule, and one base is called a nucleotide. There are only four different bases: adenine (A), thymine (T), guanine (G), and cytosine (C). They pair with each other so that A is always joined to T and G is always joined to C. Thus, the sequence of bases on one side of the ladder (for example, AGCGT) is complementary to, and determines, the sequence on the other side (TCGCA). This is the "genetic alphabet"—a small set of "letters" with which, as with the ABC's, an enormous number of messages can be written.

As might be expected, the nucleus is constantly active. Before cell division, all of the information contained within the DNA must be duplicated in a process called replication. The speed with which replication occurs is astonishing. For example, before a single *Escherichia coli* (a common intestinal bacterium, abbreviated *E. coli*) splits in two, which it does every 20 minutes, the 360,000 turns of its DNA helix must first be unwound. Next, each of the 3.6 million nucleotides on one side of the DNA molecule pulls away from its mate. As the molecule unzips, each half serves as a mold, or template, for a new



The nucleus, where the genetic material DNA is stored, is the cell's largest organelle. It is surrounded by a double membrane that is perforated with "gates" called nuclear pores, which are the routes by which genetic messages pass into the cytoplasm. The nucleolus is the site of ribosome manufacture.



Protein production begins in the nucleus at the DNA (a). A coded message for a protein leaves the nucleus in the form of RNA (b) and goes to either free ribosomes (c) or to ribosomes bound to the endoplasmic reticulum (d). When released from a free ribosome, a protein (e) can become incorporated into cytoskeletal fibers (f) or into such organelles as a mitochondrion (g) or a peroxisome (h). Proteins made in the endoplasmic reticulum leave in a vesicle (i) and migrate to the Golgi apparatus (j). Proteins are sorted in the Golgi and are then carried in vesicles to lysosomes (k), or are secreted (l) or incorporated into the cell's surface membrane (m).

molecule. In a matter of minutes, a total of 7.2 million free nucleotides are brought to each template and attached A to T and G to C. Finally, each new double strand retwists itself into a helix. (In prokaryotes, such as *E. coli*, the DNA exists as a large, single molecule rather than as multiple chromosomes.)

All of this molecular maneuvering must be performed both rapidly and accurately. If nucleotides are lost, rearranged, or erroneously paired, the garbled instructions that result could lead to a nonfunctioning protein when the DNA's code is translated.

After replication in human cells, the DNA condenses into 46 pairs of chromosomes. At this point, the membrane surrounding the nucleus breaks down, the chromosome pairs pull apart, and one member of each pair moves to the opposite pole of the cell. Then the cell divides, forming two identical daughter cells, each with 46 chromosomes. The production of sperm and egg cells is a more complex process in which a second division of the nucleus occurs, resulting in cells that have 23 chromosomes each, instead of 46. When an egg is fertilized by a sperm, the full complement of 46 chromosomes is restored.

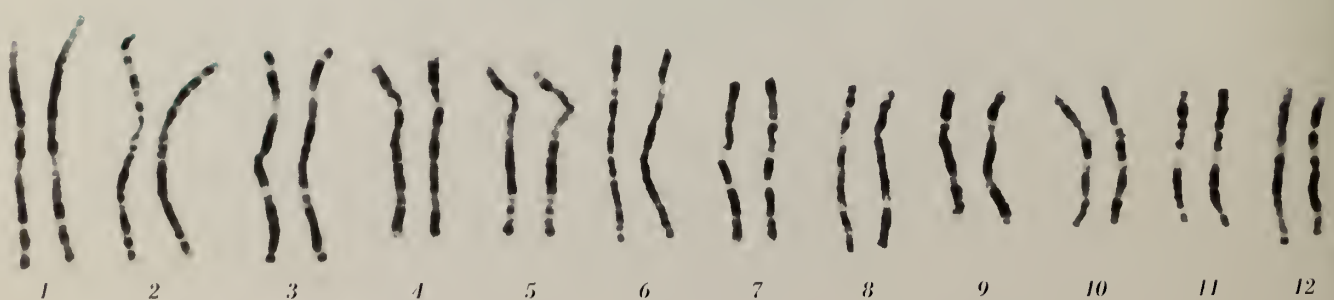
Each of us begins as a single, fertilized cell, a microscopic package that contains within its DNA the directions for everything that we can become. The single cell then divides again and again. Each new cell contains the same kinds of molecules and even the same amount of water as the "parent" cell.

Some of our cells are very short-lived and are repeatedly replaced. Scavenger white blood cells, for example, circulate and consume invading particles for only a few days before they die. In contrast, our brain cells never reproduce. Most live as long as we do, but when one dies, it is not replaced.

At any instant, only certain genes in a cell are "on," or "expressed," and giving orders for the production of specific proteins. Some of the instructions that switch these genes on or off are generated as a result of interactions between the surface membrane and the environment. Thus, the commands from the nucleus are influenced by what goes on outside the cell as well as by the cell's genetic program.

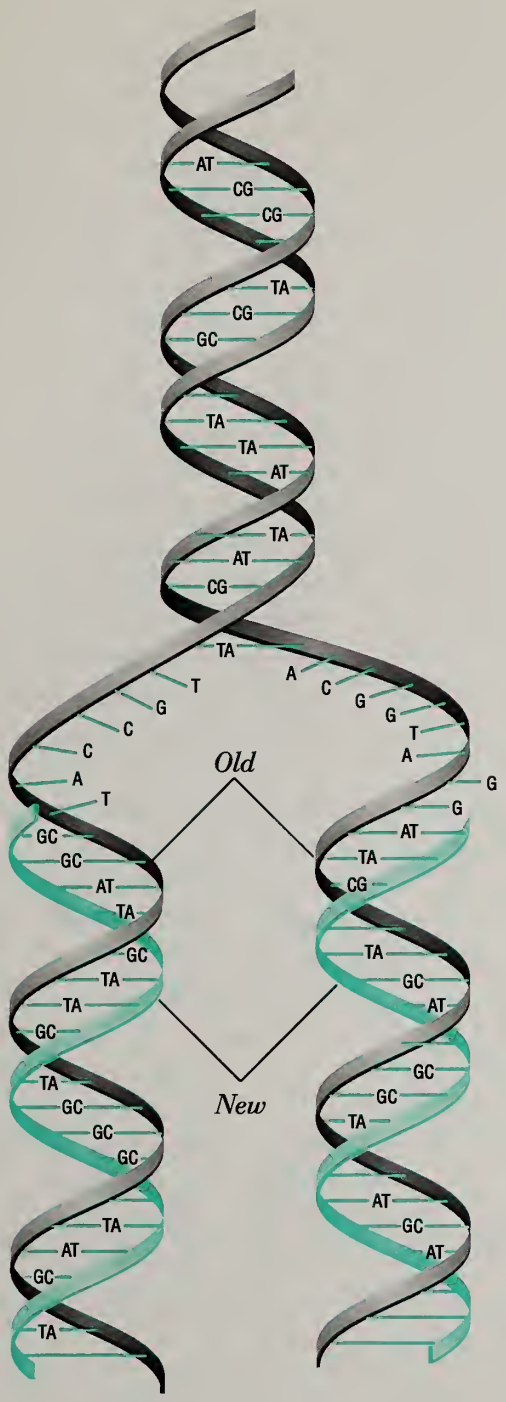
An order to make a protein begins when the appropriate genes are transcribed from the DNA into strands of another kind of nucleic acid, called messenger ribonucleic acid (mRNA). Messenger RNA is manufactured by transcribing just one chain of

Chromosomes of a normal human male



the DNA double helix (one side of the twisted ladder). A strand of mRNA is complementary to the DNA from which it is transcribed, except that each adenine of the DNA is paired with a uracil (U), instead of with a thymine. For example, the stretch of DNA bases ATCG is transcribed into the mRNA sequence UAGC.

After additional processing, the mRNA leaves the nucleus through pores in the nuclear membrane and carries its message into the cytoplasm, while the DNA remains safely in the nucleus. Once in the cytoplasm, the messenger RNA moves to tiny organelles called ribosomes, the factories in which the next step of protein manufacture, called translation, takes place. ■



To replicate before cell division, the DNA double helix separates and unwinds and each strand acts as a template for the formation of a mirror image according to the rules of base pairing: A with T, and G with C. This results in two daughter DNA molecules whose sequences are identical to those of the original DNA.

Proteins, Workhorses of the Cell

Proteins are intensely studied by biomedical researchers because these molecules are involved in nearly every biological function. Proteins include the enzymes that allow the chemical reactions necessary to life to take place efficiently. They also include many of the hormones that regulate growth and development. They are important components of the cell's physical structure, making up half of the cell's dry weight. They help transmit messages from nerve cells and work in muscle cells to convert chemical energy into mechanical energy, which permits movement. Proteins in the cell membrane control molecules entering and leaving the cell. Hemoglobin proteins in red blood cells transport oxygen through the bloodstream, and antibody proteins fight infection.

Proteins are made of strings of amino acids ordered according to instructions contained in the DNA. A protein's primary structure is this linear chain of amino acids; however, almost immediately after it is created the chain springs into helices, sheets, or other shapes that form the protein's secondary structure. The shapes then fold and coil further into a complex, three-dimensional structure. This is the active form of the protein that can bind to, and interact with, other molecules.

The many proteins that are enzymes act as catalysts, speeding up reactions without being permanently altered themselves. Without enzymes, many

cellular processes would proceed slowly or not at all. According to chemist Ronald Breslow of Columbia University, enzymes work so well that a process that takes 5 seconds (such as reading this sentence) with enzymes would take 1,500 years without them. Enzymes can do their jobs—often, cutting apart or splicing together other molecules—over and over. They also have great specificity. Like a lock, each enzyme will accept only appropriately shaped "keys" (called substrates). Each cell contains thousands of kinds of enzymes.

Once a protein is purified, the sequence in which the amino acids occur on the chain can be determined. Knowing the amino acid sequence of a protein is extremely important for many reasons. For example, it may help scientists synthesize large quantities of the protein for research or commercial purposes. Direct sequencing of a protein, although much quicker and more accurate now due to the development of automated equipment, is an enormously time-consuming task. It is technically easier to sequence the DNA of the gene that codes for the protein. For this reason, in most cases protein sequencing has been replaced by isolating, cloning, and sequencing a gene by recombinant DNA techniques. The protein sequence is then inferred from the DNA sequence that codes for it.

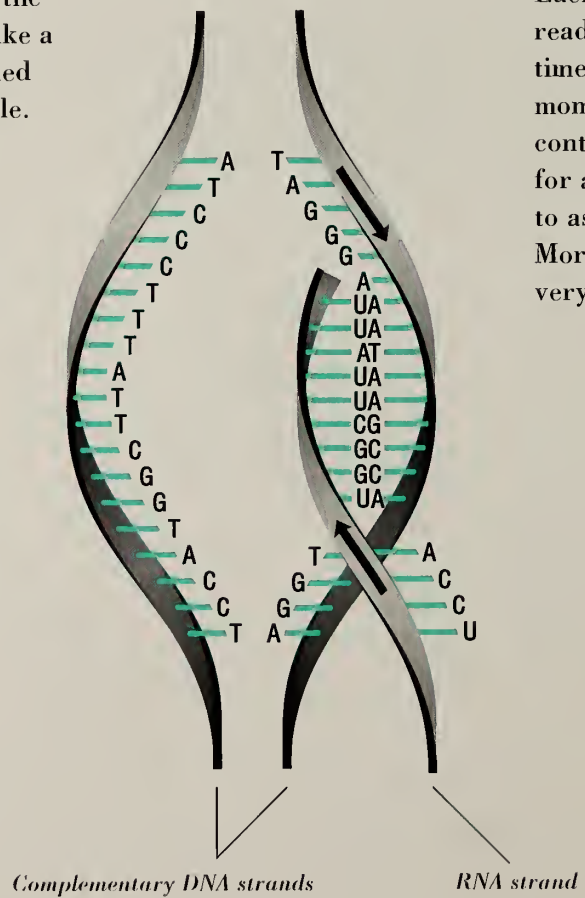
Even if the sequence of amino acids in a protein is known, scientists cannot predict how the protein will fold into its final, active shape. Many researchers are working to solve this so-called "folding problem." They feel that if they can learn the rules by which proteins fold, it will open the way to synthesizing engineered, artificial proteins with many therapeutic, industrial, and manufacturing applications. ■

Ribosomes, which were discovered in the mid-1950's, are extremely tiny—less than 30 nanometers in diameter. However, due to their crucial role in protein manufacture, ribosomes can also be extremely numerous. In *E. coli*, for example, ribosomes account for one-fourth of the cell's mass. A ribosome is made of two unequally sized subunits, each of which is composed of at least 40 different proteins and a form of RNA called ribosomal RNA.

During translation, a strand of mRNA moves between the two parts of a ribosome like a piece of thread being pulled through the eye of a needle. The ribosome reads the message of the mRNA not one nucleotide at a time, but rather in groups of three. These groups, called codons, are like words. Each word specifies one of the 20 different amino acid subunits of a protein or is a signal to start or stop making a protein. For example, the codon AGC in mRNA is translated into the amino acid serine, whereas nucleotides in a different order, say GCA, code for alanine.

The amino acids called for by the mRNA are brought from the cytoplasm to the ribosome by a third kind of RNA, transfer RNA (tRNA). This small molecule is a connector: One end carries three nucleotides, known as the anticodon, which will join to a codon in the mRNA according to the rules of base pairing (A with U, and G with C). The molecule's other end carries an amino acid. As the mRNA passes through the ribosome, tRNA brings the correct amino acids in and they are linked together by chemical bonds to form a long chain. When all the amino acids for a protein are joined, the chain is released.

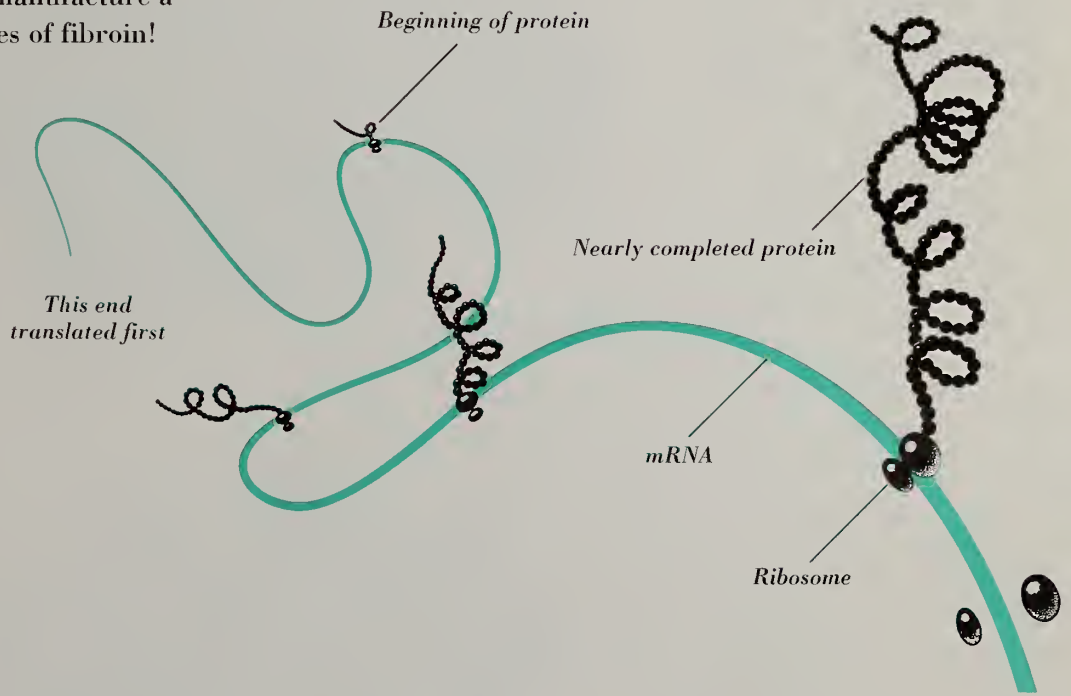
Each strand of mRNA can be read many thousands of times. Indeed, at any one moment a strand of mRNA containing the instructions for a protein may be attached to as many as 30 ribosomes. Moreover, ribosomes work very quickly to connect the



required amino acids into a protein. Each ribosome in a single *E. coli*, for example, can link 15 amino acids in a second. The speed and efficiency of translation means that each gene is capable of directing the manufacture of very large quantities of protein. For instance, in each cell of a silkworm's silk gland there is a single gene that codes for the protein fibroin, the chief component of silk. Each time it is activated, the gene can make 10,000 copies of its specific mRNA, and *each* copy of mRNA can direct the synthesis of 100,000 molecules of fibroin. In 4 days, a silk gland cell can manufacture a billion molecules of fibroin!

Ribosomes fall into two categories: those that are free in the cytoplasm and those that are bound to membranes. The two kinds of ribosomes play similar roles in the manufacture of proteins. But while the free ribosomes leave the proteins equally free to float in the cytoplasm, the bound ribosomes transfer their finished proteins into a large, cobwebby organelle—the endoplasmic reticulum. ■

In the nucleus, DNA's instructions are transcribed (below left) into a messenger molecule of ribonucleic acid (RNA). The code in a strand of messenger RNA is translated into a protein (below right) in tiny organelles, called ribosomes, in the cytoplasm.



In 1945, as the electron microscope was becoming a useful research tool, Albert Claude of Belgium and Keith Porter, who was then at The Rockefeller Institute, used it to discover a vast network of channels bounded by membranes in the cytoplasm of chick embryo cells. At times, this network looked like the concentric circles of a slice of onion. Porter called this network the endoplasmic reticulum (ER) because it was more concentrated in the inner (endoplasmic) region of the cell than in the peripheral (ectoplasmic) region. Similar networks were later found in all eukaryotic cells, except mammalian red blood cells.

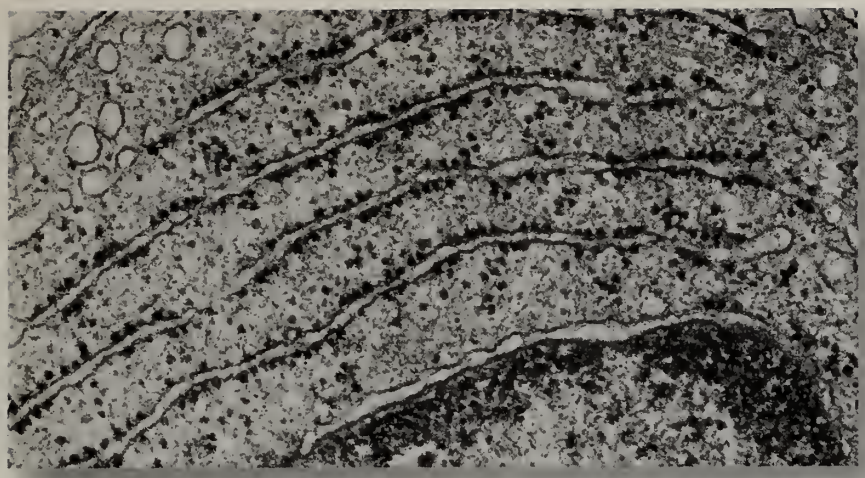
It was discovered that the membranes of the endoplasmic reticulum all interconnect, forming a system of tubes and flattened sacs. Some parts of the endoplasmic reticulum look smooth, while others appear rough because they are dotted with ribosomes that form granules on their outer surfaces.

The smooth endoplasmic reticulum is involved in the synthesis of fatty acids and membrane components. This ER also contains enzymes that help detoxify and process chemicals. It is especially prevalent in liver cells.

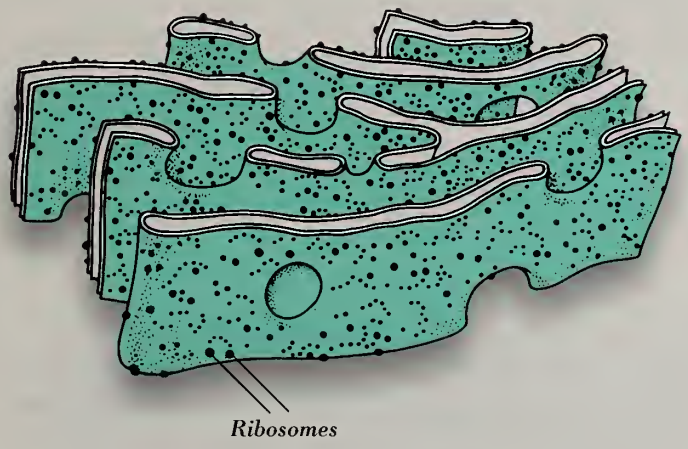
Many of the proteins the ribosomes of the rough endoplasmic reticulum synthesize are intended to be exported outside the cell. These proteins carry specific amino acid sequences, or "addresses," that allow them to enter the inner space of the rough endoplasmic reticulum, where they undergo additional biochemical modifications.

In the mid-1950's, George Palade, then of The Rockefeller Institute, concluded that the amount of rough endoplasmic reticulum in a cell corresponds closely to the quantity of protein the cell exports. For example, white blood cells that produce infection-fighting immune system proteins called antibodies have highly developed rough endoplasmic reticula.

Most of the proteins leaving the endoplasmic reticulum are still not mature; they must undergo further processing in another organelle, the Golgi apparatus, before they are ready to perform their functions within or outside the cell. ■



Electron micrograph shows the folds of the endoplasmic reticulum thickly dotted with tiny dark bodies, the ribosomes.

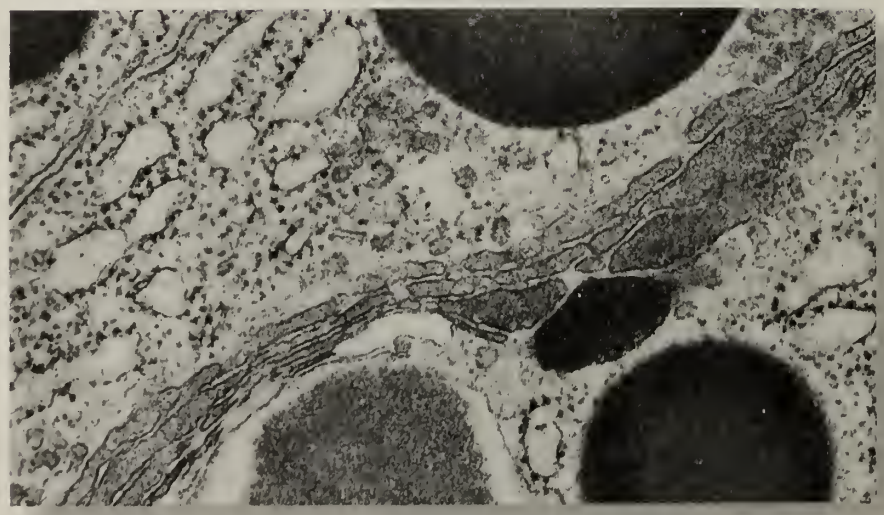


Drawings show both the ribosome-covered rough endoplasmic reticulum (top) and the ribosome-free smooth endoplasmic reticulum (bottom).

In 1898, the Italian scientist Camillo Golgi, who had been studying stained owl and cat nerve cells under his light microscope, saw a cell structure that did not look like the nucleus. Although some biologists at the time thought the structure might be an artificial one, perhaps related to the stains that Golgi had used, he believed that the newly found organelle played a role in protein secretion.

In the 1960's, Palade and his colleagues confirmed Golgi's theory by using radioactive labeling, staining, and electron microscopy to follow proteins in pancreatic cells as they moved from the rough endoplasmic reticulum, through the Golgi apparatus, and into the secretory granules that carried them out of the cell.

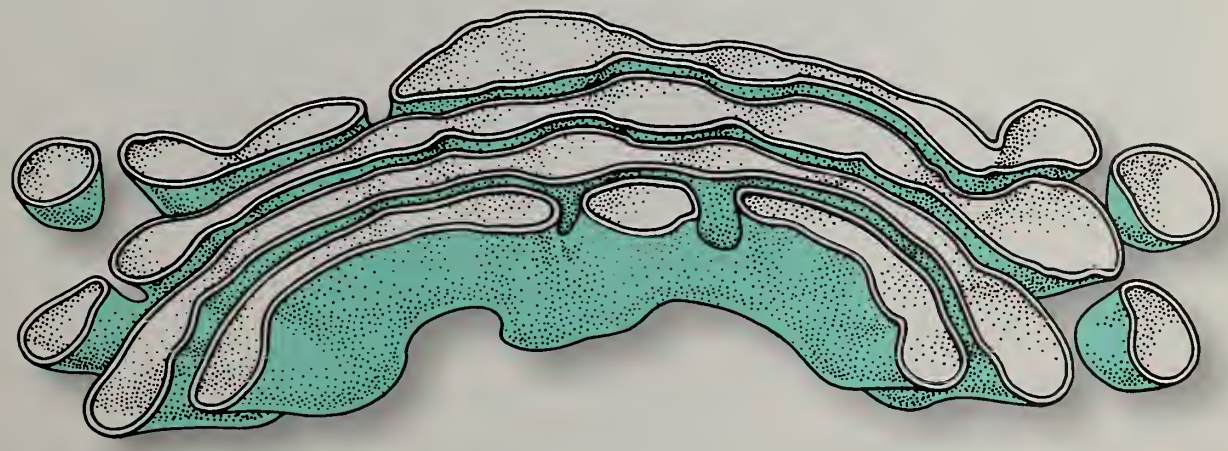
It is now known that each Golgi apparatus consists of a stack of flat, membranous sacs that are piled one on top of the other like dinner plates. The stack is composed of three distinct regions, and each sac in the organelle contains enzymes that modify proteins as they pass through. The sacs closest to the nucleus receive vesicles (membrane-bound sacs) filled with protein molecules from the endoplasmic reticulum. The proteins must pass through all the sacs in sequence to be processed correctly.



The Golgi apparatus plays an important role in transforming many newly made proteins into mature, functional ones. Moreover, the Golgi apparatus serves to "package" certain proteins, including enzymes and hormones, into vesicles that will later be secreted from the cell. And finally, the Golgi apparatus adds "addresses" to proteins that are destined to go to another organelle, called the lysosome.

The elaborate organization of the Golgi apparatus into separate compartments prevents the release of thousands of enzymes that, if mixed, would result in uncontrolled biochemical reactions inside the cell. ■

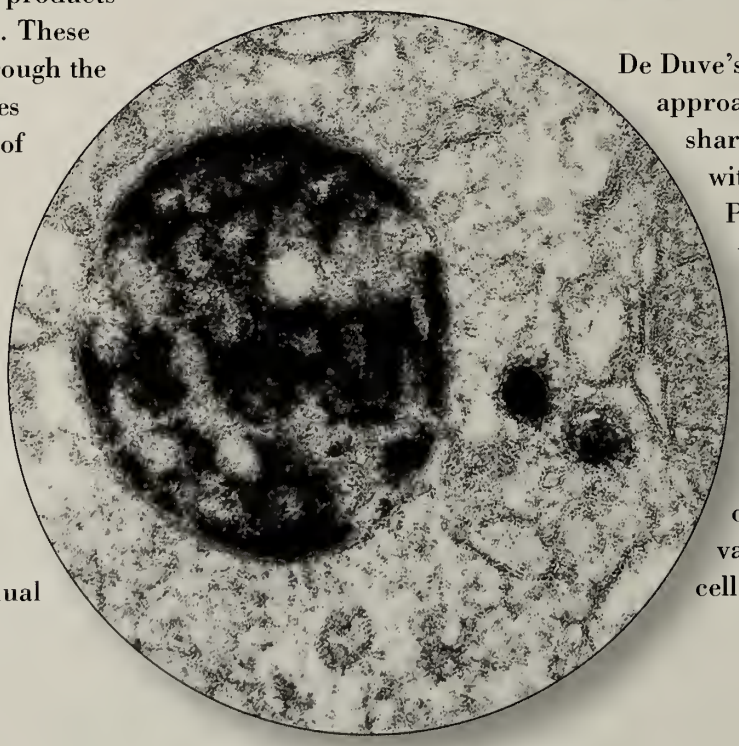
Electron micrograph (opposite page) and drawing (below) show a series of cup-shaped sacs making up a Golgi apparatus.



When a white blood cell engulfs a bacterium and destroys it, the white blood cell's lysosomes do most of the work. They fuse with the vesicle of engulfed material and release digestive enzymes to break up the material. Similarly, when a cell takes in large molecules of food, enzymes in the lysosomes break the food down into smaller and simpler products that the cell can use. These products diffuse through the lysosomes' membranes and go into the rest of the cell, where they serve as building blocks for various structures, until nothing is left inside the lysosomes but indigestible material, and the lysosomes become what are called residual bodies. In some cells, the residual bodies then migrate to the cell surface and eject the undigested material into the external environment.

Lysosomes were discovered by a Belgian researcher, Christian de Duve, in 1949, when he homogenized some animal cells and separated them into various components by using an ultracentrifuge. After one of these components had been left standing for a few days, de Duve noticed that the level of a certain enzyme in it had risen dramatically. Since this enzyme had not attacked any part of the cells before they were ground up, he reasoned that it must have been kept segregated within the cell—probably inside some kind of organelle. He also knew that he had used a relatively gentle method of homogenization, which could have allowed the unknown organelle to remain intact. Presumably, it released its contents later.

De Duve's biochemical approach, for which he shared the Nobel Prize with Claude and Palade in 1974, was soon supplemented by electron microscopy. But it proved difficult to identify the new particles, since, unlike other organelles, lysosomes vary in shape from cell to cell. Finally, in



An electron micrograph showing two small lysosomes and one large lysosome. These organelles contain enzymes capable of breaking down various substances.

1955, Alex Novikoff of the Albert Einstein College of Medicine clearly identified some lysosomes in rat liver cells, and it is now known that lysosomes (whose name refers to the fact that their enzymes can lyse, or digest, substances) exist in all eukaryotic cells. In fact, lysosomes contain over 40 different enzymes that can digest almost anything in the cell, including proteins, RNA, DNA, and carbohydrates.

At about the same time that de Duve and his colleagues were describing the biochemistry of lysosomes, they detected another enzyme-containing organelle. In 1965, de Duve proposed that the organelle be called a peroxisome because it appeared to both generate and break down hydrogen peroxide, a corrosive molecule composed of two atoms each of hydrogen and oxygen.

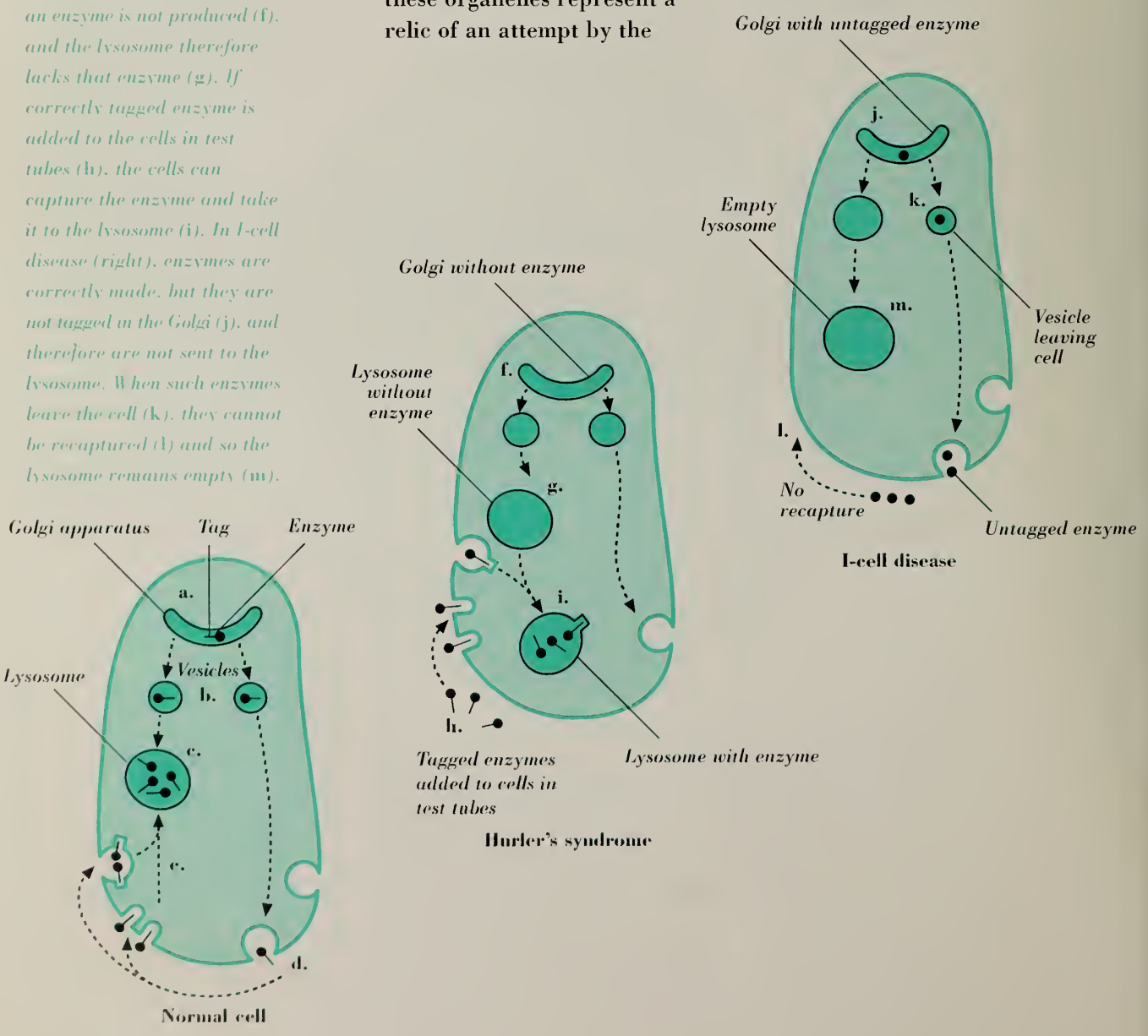
Today it is known that peroxisomes exist in most eukaryotic cells, and that they are especially prominent in mammalian liver cells. The membrane that surrounds a peroxisome is usually permeable, permitting many small molecules to enter easily. Peroxisomal enzymes remove hydrogen atoms from these small molecules and join the hydrogen to atoms of oxygen to form hydrogen peroxide. One of the peroxisomal enzymes, catalase, then neutralizes the hydrogen peroxide by catalyzing its breakdown into water and oxygen. This two-step process is the method that peroxisomes in the liver use to break down molecules of alcohol into substances that can be eliminated from the body. About one-quarter of the alcohol that enters the liver is processed in peroxisomes.

In his early descriptions of peroxisomes, de Duve called them "fossil organelles" because of their primitive nature and seemingly expendable actions. (All of the enzymes found in peroxisomes are also found

The sorting of lysosomal enzymes in a normal cell is shown below. First, enzymes receive a chemical "address label" in the Golgi apparatus (a) and move in vesicles (b) to a lysosome (c). Any enzyme that is accidentally swept out of the cell (d) is recaptured and taken back to the lysosome (e). If some part of the process goes awry, a lysosomal storage disorder can result. For example, in Hurler's syndrome (center), an enzyme is not produced (f), and the lysosome therefore lacks that enzyme (g). If correctly tagged enzyme is added to the cells in test tubes (h), the cells can capture the enzyme and take it to the lysosome (i). In I-cell disease (right), enzymes are correctly made, but they are not tagged in the Golgi (j), and therefore are not sent to the lysosome. When such enzymes leave the cell (k), they cannot be recaptured (l) and so the lysosome remains empty (m).

elsewhere in the cell.) However, it is now known that a rare, fatal genetic disorder called Zellweger's syndrome is the result of malformed peroxisomes, indicating that peroxisomes do have a vital role in the healthy cell.

The ability of peroxisomes to use oxygen in chemical reactions has led many scientists to conjecture that these organelles represent a relic of an attempt by the precursors of eukaryotic cells to "cope" with oxygen as it accumulated in the prehistoric atmosphere. Peroxisomes cannot, however, couple oxygen use with energy production. That ability is restricted to mitochondria—the "energy converters" of eukaryotic cells. ■



Lysosomes in Health and Disease

If the cell does not produce a certain lysosomal enzyme or if an enzyme is not properly "addressed" in the Golgi apparatus, a lysosomal storage disease can result. These diseases are caused by a massive accumulation of material that should have been digested in the lysosome. Persons with the lysosomal storage disease known as Hurler's syndrome, for example, cannot break down large molecules of sugar compounds called glycosaminoglycans because their lysosomes do not contain the enzyme iduronidase. Glycosaminoglycans accumulate in the lysosomes, swelling them so much that the functioning of the entire cell is impaired.

A particularly severe lysosomal disorder is known as I-cell disease. Children born with this disease lack the entire range of lysosomal enzymes. The enzymes are made, but they are dumped outside the cell instead of being sent to the lysosomes. Various cellular nutrients thus cannot be digested and so pile up in dark lumps, called inclusion bodies, within the lysosomes. The disease affects the kidneys, heart, and nervous system, and children with it usually die of heart failure or pneumonia before reaching puberty.

In the early 1970's, Elizabeth Neufeld, who was then at the National Institutes of Health, showed that the lysosomal enzymes of persons with I-cell disease emerge from the Golgi apparatus without the chemical tag they need to be directed to the lysosomes. She also showed that the defect could be corrected in test-tube cultures of cells taken from people with the disease. The corrective factors she supplied were the specific, properly tagged enzymes that the cells lacked.

Although enzyme replacement therapy is not being used to treat people with lysosomal storage diseases like Hurler's syndrome, it is being used to treat a different group of disorders called lipid storage diseases. Enzyme replacement therapy for many disorders presents challenges to researchers, however, because purified enzymes injected directly into the body tend to be quickly destroyed or inactivated. It is particularly difficult to get enzymes into brain cells—an important problem now under investigation, since several lysosomal storage diseases produce severe mental retardation. ■

One and a half billion years ago, scientists believe, eukaryotic cells derived the energy they needed through a variety of relatively inefficient processes, none of which required oxygen. Oxygen, a waste product of some of these processes, gradually began to accumulate in the atmosphere. It was at this time, scientists hypothesize, that a primitive eukaryotic cell engulfed a primitive bacterium that had acquired the ability to use oxygen to produce large quantities of energy. Over the eons, a symbiotic relationship evolved between the cells, and today almost all plant and animal cells have organelles that are the descendants of the primordial energy producers. In animal cells, these organelles are called mitochondria. Plant cells have both mitochondria and a second kind of energy-producing organelle, the chloroplast.

Chloroplasts use the energy in sunlight to convert molecules of carbon dioxide and water into molecules of sugar, a form of energy that can be stored in the plant cell. (Oxygen is given off as a byproduct of this process, which is called photosynthesis.) When an animal eats a plant (or another animal that has itself eaten plants), the plant's sugars are broken back down into carbon dioxide and water, with the help of oxygen and an arsenal of enzymes, releasing large amounts of stored energy. This energy is immediately converted to yet another form—molecules of adenosine triphosphate (ATP).

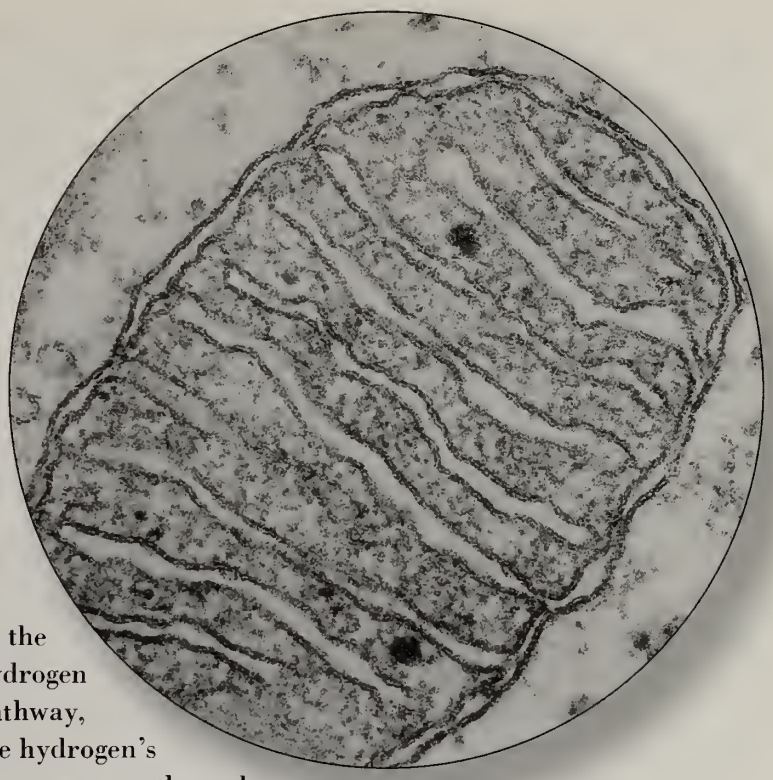
ATP is often called the universal currency of cellular energy. It is a convenient way for cells to store the energy they need for such processes as protein manufacture, DNA replication, and the construction of new organelles. ATP is also required for such mechanical work as muscle contraction, pumping water through membranes, and cell movement. Following the first stages of sugar breakdown, the complicated process of energy transfer from sugar to ATP takes place within the animal cell's mitochondria.

Besides supplying energy, mitochondria help to control the concentration of calcium and other electrically charged particles in the cytoplasm. They also break down and recycle the energy contained in fatty acids and amino acids.

Mitochondria are the largest organelles in an animal cell, after the nucleus, yet some cells have more than a thousand of them. They vary in diameter from 0.5 to 1 micrometer and in length up to 7 micrometers, and can be seen with a good light microscope. Mitochondria are usually represented as oval-shaped, but in life they can change shape quite readily. They swell or contract in response to various hormones and drugs and during ATP

manufacture. This swelling and contracting appears related to the movement of water through cells, and is particularly evident in the kidneys, through which 180 liters of blood are filtered daily.

Although mitochondria were first observed in the 1880's, it took many years for scientists to understand the organelles' function. The process by which mitochondria use oxygen to release the chemical energy stored in food is called cellular respiration. In the early 1900's, it was discovered that the biochemical reactions of this type of respiration fall into two main groups: the carbon pathway, in which sugar is broken down into carbon dioxide and hydrogen; and the hydrogen pathway, which transfers hydrogen to oxygen in stages, forming water and releasing energy.



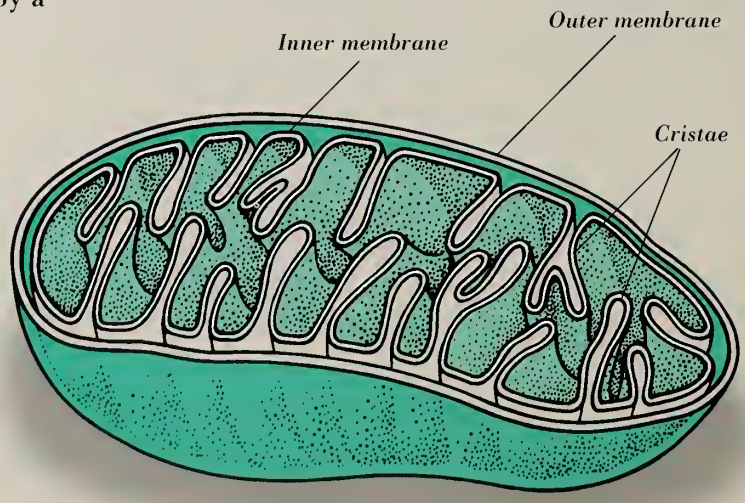
Electron micrograph showing one of the cell's many mitochondria, the organelles that convert energy from food into a form that can be stored

In the hydrogen pathway, the hydrogen's electrons pass through an "electron transport chain" made up of enzymes. As they move from enzyme to enzyme, the electrons give up part of their energy. This energy is then stored in molecules of ATP. In the end, 38 molecules of ATP are formed for every molecule of sugar that is used up in respiration.

Mitochondria are marvelously efficient at converting the chemical energy of sugar into ATP. Whereas an engine would be considered very efficient if it converted 25 percent of the energy available in gasoline into mechanical work, mitochondria routinely turn 54 percent of the available energy in sugar into ATP. This efficiency is achieved, in large part, because of the mitochondria's internal structure. In the early 1950's, Palade and a Swedish scientist, Fritiöf Sjostrand, reported that mitochondria are surrounded by a membrane and that they have a system of parallel, regularly spaced inner ridges that the scientists named "cristae." It is now known that there are two membranes around a mitochondrion: an outer membrane, separated from the rest of the organelle by a fluid-filled gap; and an inner membrane that is folded inward in many places to increase its surface area, forming the cristae. This ridged surface allows the enzymes of the electron transport chain, which are attached to the cristae, to be packed more densely within each mitochondrion, thus increasing the organelle's efficiency. This general design seems to have existed unchanged from the time that mitochondria-like cells were free-living organisms.

Mitochondria have also kept other vestiges of their existence as independent organisms. For example, mitochondria "reproduce" by splitting in half, as many modern bacteria do; they are not formed by budding from existing cellular structures or built up from simple cellular constituents, as is the case for ribosomes.

More significantly, after a billion or so years of residence within "host" cells, mitochondria (and chloroplasts) still retain some of their own DNA. The amount of this non-nuclear DNA varies significantly from organism to organism. The chloroplasts of plants, for example, have five times more DNA than do the mitochondria of mammalian cells. Human mitochondrial DNA is a circular molecule



16,569 nucleotide pairs long. Although this is less than 1 percent of the total DNA in a human cell, each mitochondrion has enough DNA to code for several of its key inner membrane proteins and its own ribosomal proteins. (All of the other proteins in a mitochondrion are coded for in the nucleus, made on free ribosomes in the cytoplasm, and imported into the organelle.)

Another curious characteristic of human mitochondria is the fact that all of a person's mitochondria are descendants of those of his or her mother; no paternal mitochondria are present. This fact has proved useful to evolutionary biologists, who can study the passage of mitochondrial DNA from generation to generation while ignoring the "interfering" information contained in the nuclear DNA, which records the genetic contributions of both parents.

Scientists have long suspected that defects in mitochondrial genes could lead to inherited disease in the same way that mistakes in nuclear DNA do. This hunch was not proven until 1988, when Douglas Wallace of Emory University showed that a rare eye disease called Leber's hereditary optic neuropathy is caused by a mutation in mitochondrial DNA. The defective mitochondrial gene prevents the optic nerves from producing enough ATP and the nerves, which need huge amounts of ATP and are thus particularly sensitive to any deprivation, die. When he announced these findings, Wallace said, "We feel that these alterations [in mitochondrial DNA] may be responsible for a wide spectrum of diseases in the brain, the central nervous system, and the musculoskeletal system."

Mitochondria, chloroplasts, and the other organelles described thus far are surrounded by membranes. But cells can also contain threadlike organelles that lack membranes. These extremely fine structures serve as buttresses, highways, and movement mechanisms for the cell. ■

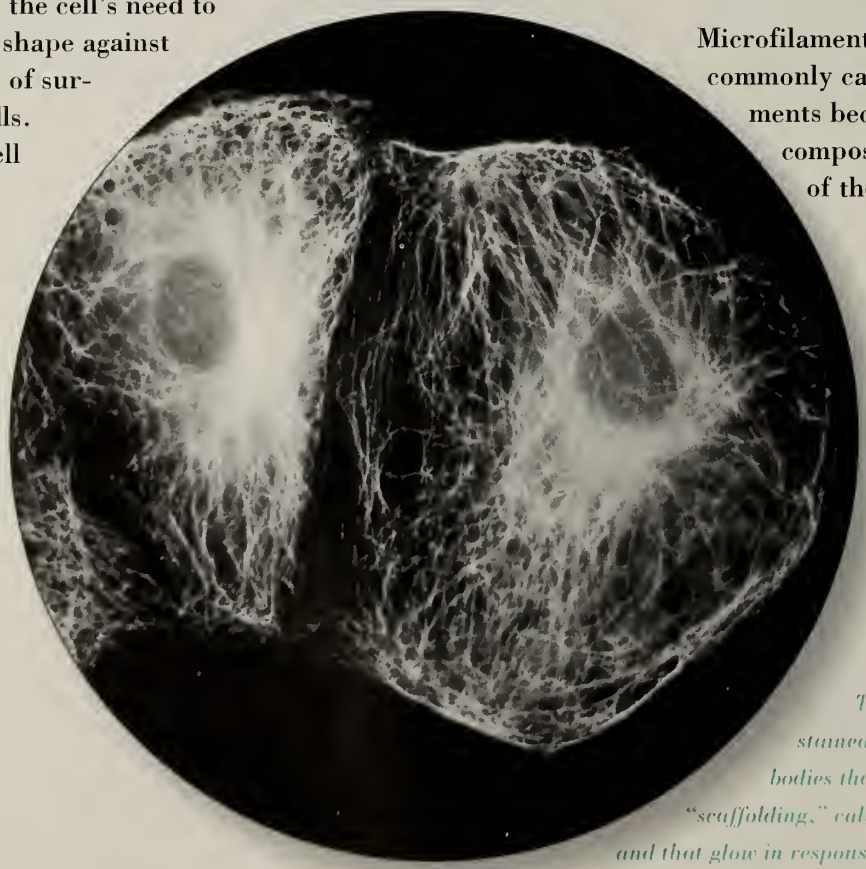
A mitochondrion is shown as if it had been sliced longitudinally. The inward folds of the inner membrane are called cristae.

Many cells in a multicellular organism must combine the seemingly contradictory traits of stability and mobility. With few exceptions, multicellular organisms begin to develop when a motile sperm meets an egg. Many cell divisions occur, and then cells migrate to their final positions. During life, individual cells divide frequently, and certain specialized cells move through the body to accomplish various tasks. In addition, every cell must have a mechanism for moving materials within itself. Balancing the need for movement is the cell's need to maintain its shape against the pressure of surrounding cells. Keeping a cell firm while enabling it to move are the twin roles played by the cytoskeleton.

For a long time, microscopists believed that the cytoplasm surrounding the cell's organelles was completely unstructured. But as scientists began to use newer and gentler fixatives to prepare cells for electron microscopy, a lacy network of fibers was revealed. These structures crisscross the cell like girders and it was hypothesized (and later shown experimentally) that, like an animal's bony skeleton, these structures play a role in giving the cell its shape and support. For this reason, they are known collectively as the cytoskeleton.

There are three main kinds of cytoskeletal fibers—microfilaments, microtubules, and intermediate filaments—which are distinguishable both by their structure and by their protein composition. All three support and stiffen the cell. In addition to their structural roles, microtubules and microfilaments are essential for a variety of dynamic, whole-cell activities, including division, contraction, and crawling, as well as for the movement of vesicles and chromosomes within the cell.

Microfilaments are more commonly called actin filaments because they are composed of "beads" of the protein actin



These cells have been stained with modified antibodies that attach to the cell's "scaffolding," called the cytoskeleton, and that glow in response to a specific wavelength of light.

arranged into long, slender chains. Each filament is only 6 nanometers in diameter; they are the thinnest of the cytoskeletal components. The role that actin filaments play in muscle contraction has been thoroughly studied over the past 40 years. In the 1950's, a British scientist, Hugh Huxley, proposed a model for muscle contraction that has since been shown to be correct. According to the model, each muscle cell comprises parallel rows of actin filaments that alternate with rows of another protein, myosin. When stimulated by an influx of calcium, projecting "arms" of myosin "grab" the adjacent actin filaments and pull, causing the muscle cell to shorten. Contraction is an ATP-requiring process; each "grab" and release by a myosin molecule uses up one molecule of ATP. In recent years, researchers have found evidence of similar actin-myosin interactions in many other kinds of cells, including cells that secrete hormones and white blood cells that move through the body to fight invading organisms.

The microfilament bundles in this skin cell have been stained with modified antibodies that glow in response to a specific wavelength of light.

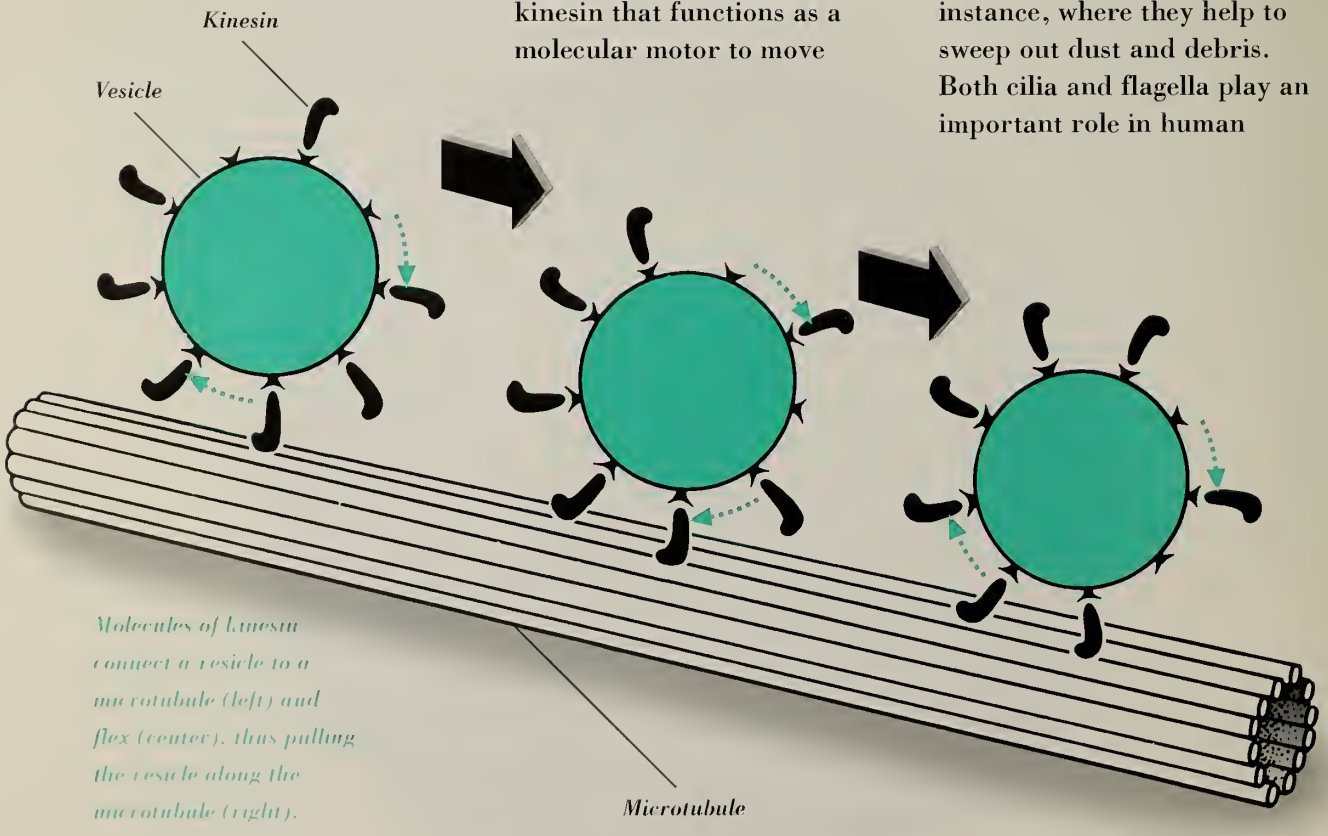
Microtubules, at 22 nanometers in diameter, are the thickest of the cytoskeletal components. They were noticed in the mid-1950's, but were seen only rarely until 1963, when the gentle fixative glutaraldehyde was developed. Each hollow tubule is composed chiefly of small, spherical subunits of proteins called tubulins. Microtubules assemble spontaneously from "pools" of tubulin when needed and, under appropriate conditions, dissolve back into their tubulin subunits. (Microfilaments also form and break down spontaneously.)

Under the microscope, microtubules can be observed growing and shrinking rapidly.

One of the most vital functions of microtubules is to aid in cell division. Just before a cell divides, small bodies called centrosomes (which are themselves composed of microtubule-like fragments) migrate to the cell's poles. An oval-shaped bundle made of microtubules forms between the centrosomes. Chromosomes attach to the bundle, which then helps to guide them to the daughter cells. In 1988, Marc Kirschner, who was then at the University of California, San Francisco, and his colleagues found strong evidence that chromosomes move toward the poles as the microtubules slowly dissolve.

Between cell divisions, microtubules act as miniature highways along which vesicles carrying such materials as hormones, neurotransmitters, and nutrients move. Using new techniques, such as video-enhanced light microscopy, scientists in several laboratories have observed microtubules interacting with a protein called kinesin that functions as a molecular motor to move vesicles and organelles along microtubule "tracks" toward the cell surface. Kinesin also moves vesicles filled with neurotransmitters along the microtubules within nerve cell axons. A second, more recently discovered motor protein, called dynein, moves vesicles in the opposite direction, toward the cell's interior.

Microtubules are also involved in the movement of cilia and flagella. These whiplike filaments project from certain cells and perform a variety of tasks. Large numbers of cilia are found on cells that line the respiratory tract, for instance, where they help to sweep out dust and debris. Both cilia and flagella play an important role in human



Molecules of kinesin connect a vesicle to a microtubule (left) and flex (center), thus pulling the vesicle along the microtubule (right).

reproduction. The coordinated beating of cilia in the oviduct produces a sort of current that draws the egg into the uterus, while the rapidly thrashing flagella of sperm help them to "swim" toward the egg.

The inherent ability of microtubules and microfilaments to assemble and disassemble rapidly allows for the construction and destruction of these cytoskeletal components to suit the needs of a moving cell. In contrast, intermediate filaments are the most stable of the cytoskeletal fibers. At 8 to 10 nanometers in diameter, they are intermediate in size between microfilaments and microtubules. Intermediate filaments are strong and are found in cells that require or provide mechanical strength, such as those of the skin and intestines. It is believed that these filaments also have other important functions in cell physiology, and researchers are studying the cause and effect of their alteration during disease.

Current investigations of the cell's organelles—the nucleus, ribosomes, endoplasmic reticulum, Golgi apparatus, lysosomes, peroxisomes, mitochondria, and cytoskeleton—hold great promise for the solution of problems in basic biology and clinical medicine. However, the key that may unlock the greatest number of health benefits may well be found in the cell's filmy membranes, particularly the surface membrane, which plays a pivotal role in maintaining the integrity of the cell and, in a larger way, in protecting the health of the organism. ■

Some microorganisms are equipped with a flagellum (composed of microtubules), which thrashes to propel the animal.

"In the beginning," wrote biologist Gerald Weissmann, "there must have been a membrane! Whatever flash of lightning there was that organized purines, pyrimidines, and amino acids into macromolecules capable of reproducing themselves, it would not have yielded cells but for the organizational trick afforded by the design of a membrane wrapping." Weissmann imagines these primitive membranes forming bubbles in which the first macromolecules were enclosed and protected from dissipation in the salty primordial seas.

A cell's outer membrane is often thought of as a boundary that distinguishes the living cell from its surroundings. And, indeed, surface membranes are crucial in keeping cells intact. Moreover, the internal membranes that wrap around many organelles in eukaryotic cells separate the cytoplasm into discrete regions, somewhat like the walls that form rooms in a house. These inner membranes enable the cell to perform many otherwise incompatible biochemical activities simultaneously, thereby greatly increasing the cell's efficiency.

Yet despite its barrier functions, the cell membrane—which is often less than 0.01 micrometer thick—is not impassive. Rather, it is exquisitely sensitive to its surroundings and selectively allows certain substances to enter and leave the cell while barring others. It takes in nutrients and excretes wastes. It sends and receives chemical and electrical messages, including signals for the cell to manufacture proteins or to divide. In multicellular organisms, it joins with other cells to form tissues.

These myriad abilities are due to the membrane's composition. Although surface membranes differ in their precise composition depending on the cell's type, and although a membrane's configuration changes from moment to moment, all membranes are composed of two basic kinds of molecules—proteins and lipids (fats).

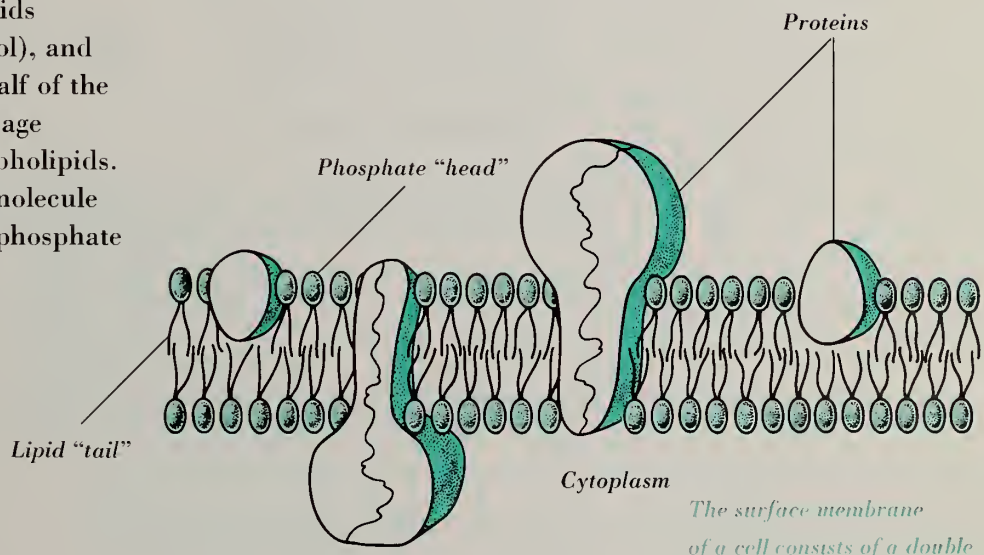
In 1972, S. Jonathan Singer and Garth Nicolson of the University of California, San Diego proposed a model to describe the relationship of proteins and lipids in an idealized membrane. They compared the proteins to "icebergs floating in a sea of lipids," and suggested that some of the proteins are folded so that the "tips" poke above and below the plane of the membrane, while the middle of the protein is embedded in the membrane itself.

Although such tripartite proteins were unknown at the time Singer and Nicolson proposed their model, they have since been shown to play important roles in a number of biological processes, including those that involve the transport of molecules into the cell. Many other membrane proteins that are attached either to the inner or outer face of the surface membrane have also been studied in detail in the years since Singer and Nicolson proposed their so-called fluid-mosaic model.

The lipids that make up the bulk of a cell's surface membrane fall into three classes: phospholipids, steroids (primarily cholesterol), and glycolipids. About half of the molecules in an average membrane are phospholipids. Each phospholipid molecule has a water-seeking phosphate "head" and two flexible, water-avoiding lipid "tails." In a surface membrane, phospholipids spontaneously arrange themselves into a double layer with the phosphate heads touching the watery interior and exterior of the cell, and the lipid tails buried in the middle of the layer.

Cholesterol is a rigid molecule that helps stabilize the membranes of animal cells. It is manufactured within the cell (in the endoplasmic reticulum) and is also brought into the cell from the blood. Cholesterol is present only in animal cells; plant cells are stiffened by a very rigid cell wall composed mainly of cellulose.

Glycolipids are composed of a sugar ("glyco" is derived from the Greek word for sweet) and a lipid portion, and make up about 5 percent of the lipid population. A person's blood group (O, A, B, or AB) is determined by the particular kind of glycolipids present on the surface of his or her red blood cells. ■



The surface membrane of a cell consists of a double layer of lipid molecules interspersed with protein molecules. Surface membranes play many roles, including keeping the cell intact and allowing appropriate substances to enter and leave the cell.

The oily lipids of a cell's surface membrane serve admirably to prevent the cell's water-soluble contents from leaking out. However, in preventing such leaks, the cell is confronted with another problem—how to transport wastes and cell products out of the cell and allow nutrients and other substances in, without either shrinking or swelling too much.

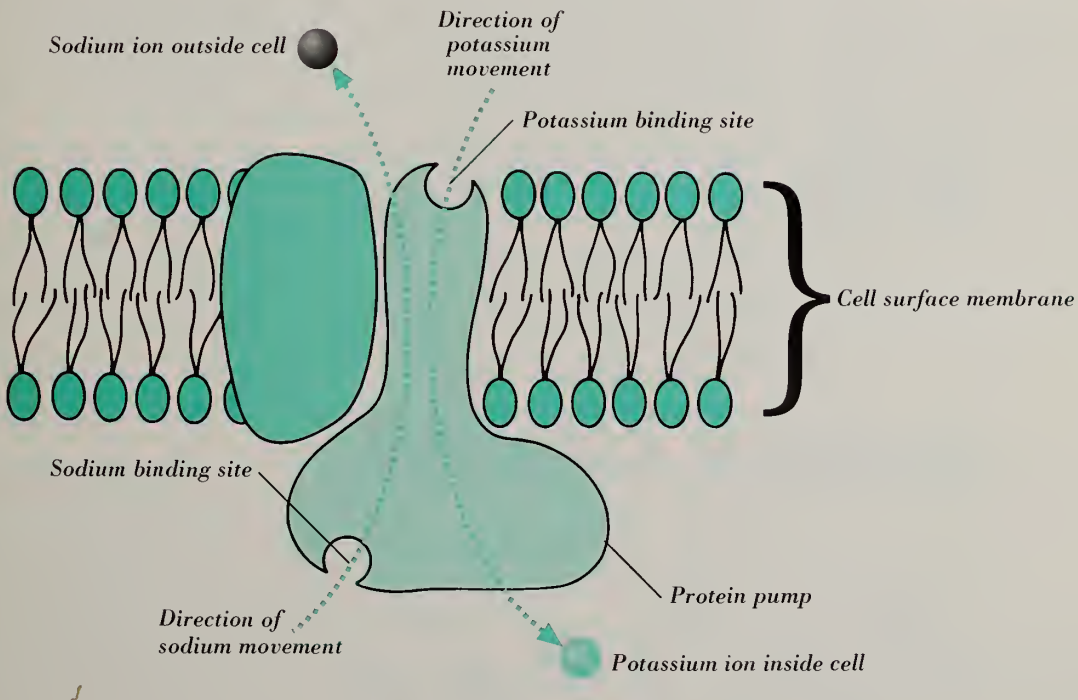
Over eons, cells have evolved a wide variety of transport mechanisms to ferry substances across the membrane. Transport may be either "passive," which requires no energy, or "active," which uses ATP. Also, a molecule may either pass directly through the membrane (usually through a pore created by a specific transmembrane protein) or it may be carried in when a bit of the surface membrane folds inward around the entering particle, then pinches off and carries the particle into the cell. The method used to import a substance depends on a combination of its size, chemical composition, electrical charge, abundance, and ability to dissolve in lipids.

Oxygen, nitrogen, and other small molecules that can dissolve easily in lipids move readily back and forth across the membrane. Importantly, because of its small size and the distribution of its electrical charge, a water molecule can also pass relatively easily through the membrane even though water is quite insoluble in oil.

In contrast, large molecules, such as proteins and sugars, cannot pass through the membrane unassisted. A variety of transport systems, many of which involve surface proteins, are used to ferry these substances into and out of the cell. Surface

membrane lipids are also highly impermeable to all electrically charged molecules, no matter how small they are. ATP-requiring protein "pumps" are employed to transport these particles, which are called ions.

One well-studied pump system is the sodium-potassium pump. This membrane protein consumes more than a third of the cell's total ATP production in an endless cycle of pumping sodium ions out of the cell while drawing potassium ions in. The difference in ion concentration inside and outside the cell creates a source of potential energy that can be used for a variety of tasks, including the propagation of electrical signals in nerve cells. ■



The unique characteristics of a cell depend, in large measure, on what kinds of receptor proteins it has. Like a lock that accepts only an appropriately shaped key, each different receptor will function only when the correctly shaped blood-borne molecule (called a ligand) attaches to it.

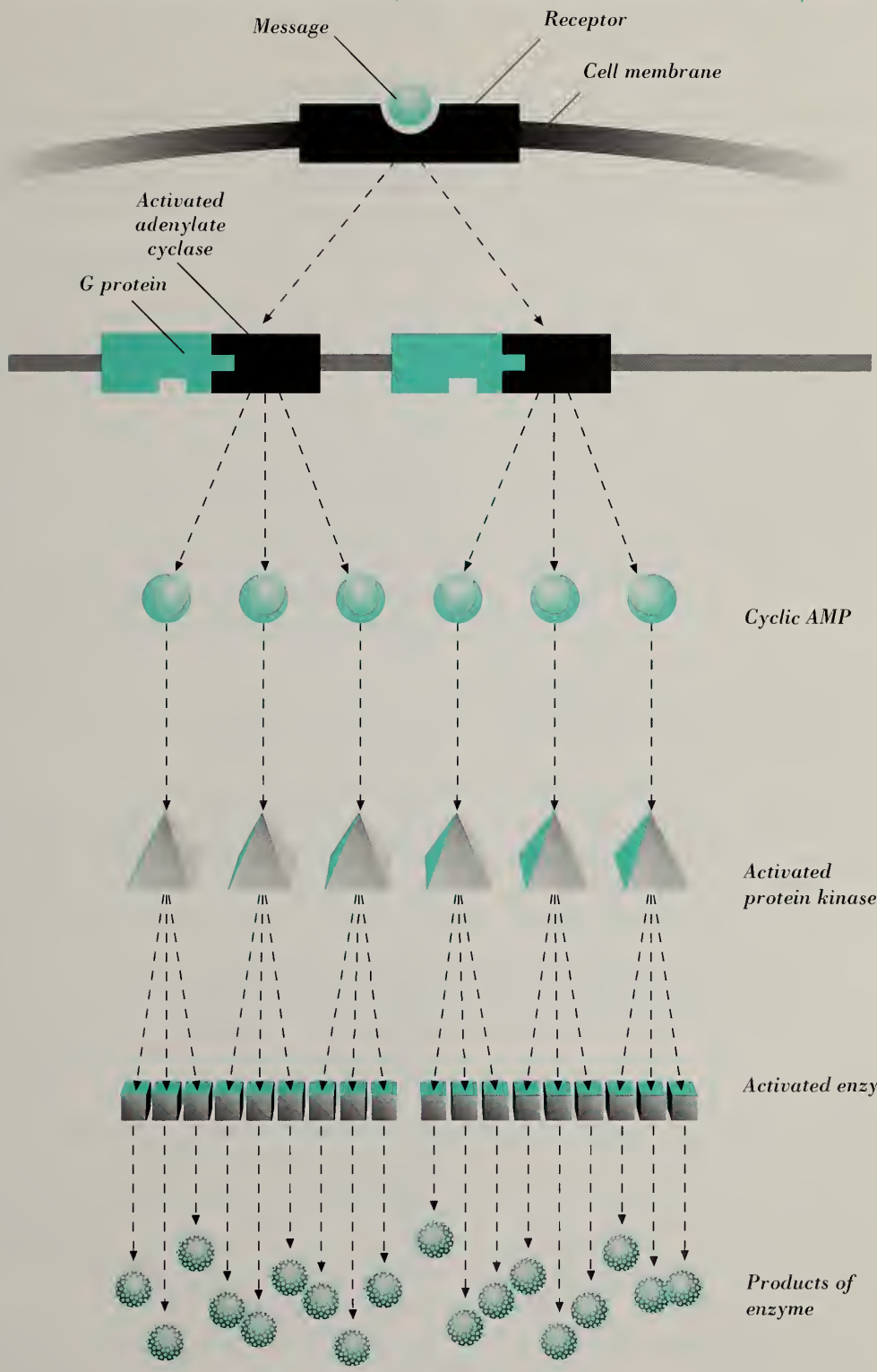
Many hormones exert their effects through receptor proteins that transfer the signal and generate "second messengers" within the cell. One of the best understood of these second messenger systems employs proteins called G proteins.

In the G protein system, when a "first messenger" (such as a hormone) reaches the cell surface, it binds to a receptor that then sends a signal to a G protein located on the interior side of the cell membrane. Depending on its type, the activated G protein then either stimulates or inhibits the activity of any of a number of enzymes, including one called adenylate cyclase. Stimulating this enzyme causes cyclic AMP, a common second messenger, to be produced. Cyclic AMP then sets off a chain reaction that eventually results in changes in the shapes of certain proteins in the cell, which, in turn, lead to still other cellular responses. When levels of the first messenger drop, the G protein "switches off" and the response terminates.

The cell appears to employ this complex signaling system because it increases both the efficiency and speed of message transmission. A single incoming messenger molecule triggers a cascade of reactions that eventually results in a large amplification of the original message. Furthermore, the time elapsed between the arrival of a signal at a G protein and a cellular response is often only a few fractions of a second. For example, light-sensitive eye cells respond to as little as one photon of light in just a few milliseconds through a G protein-mediated system. In contrast, other cells take as long as 30 seconds to respond to signals from the environment.

Certain diseases impair the functioning of the second messenger system and cause profound cellular malfunction. A toxin produced by the organism that causes cholera, for example, "locks" the G proteins of intestinal cells into the "on" position so that they are constantly stimulating the production of cyclic AMP. This causes vast amounts of fluid to cross the lining of the gut, causing the often-fatal diarrhea associated with cholera. ■

How a signal brought to the cell by a single message molecule is amplified through the second messenger system.



A message molecule binds to a receptor and activates it.

Each activated receptor activates many G proteins (two shown), which each activate one molecule of the enzyme adenylate cyclase.

Using ATP, each molecule of adenylate cyclase makes many molecules of cyclic AMP.

Each cyclic AMP molecule activates one molecule of an enzyme called protein kinase.

Each protein kinase molecule activates many molecules of an enzyme.

Each copy of the activated enzyme carries out its function.

A Breakdown in the LDL System Can Now Be Treated

The low-density lipoprotein (LDL) system works like a thermostat to ensure that the cell always has enough cholesterol for life and growth, without accumulating too much of it. Because of a defective gene, however, the cells of people with familial hypercholesterolemia (FH) are severely deficient in LDL receptors. Their livers manufacture cholesterol and release it into the blood, but since their cells do not have enough LDL receptors, not enough cholesterol is taken up. Instead, it begins to clog their arteries.

People who have two genes for FH suffer from heart disease from a very early age, sometimes as early as age 1. Persons with only one defective gene do make some LDL receptors, but still have a high risk of heart attacks, and often have an attack before age 30. FH is a relatively common genetic disorder; as many as 1 person in 500 carries one copy of the defective gene.

Michael Brown and Joseph Goldstein of the University of Texas Health Science Center at Dallas determined both the precise genetic defects and the role of the LDL receptor in FH. In 1985, both men were awarded Nobel Prizes for this work. Their studies also led to the development of lovastatin, a drug to treat FH. Lovastatin has a dual action: It reduces the liver's ability to manufacture cholesterol so that fewer LDL's enter the bloodstream, and it causes the LDL receptors that the person has to take up cholesterol more efficiently.

The mechanism Brown and Goldstein outlined for LDL receptors is but one example of the process of receptor-mediated membrane transport. The cell also uses protein receptors to bind or take in such substances as insulin, transferrin (an iron-bearing compound), and cell complexes that are produced when the immune system is activated. ■

We are deeply indebted to the scientists of the past who first revealed the marvels of the cell. As productive as the past has been, however, the future promises to be still more exciting as researchers gain an even greater understanding of cell activities and apply that understanding to questions of health and disease.

In the three centuries since Robert Hooke turned his microscope on bits of dried cork, much has been learned about the world inside the cell. Directed by the genes and influenced by the environment, cells perform an astonishing array of tasks and take on a variety of forms suited to their work. Cell biologists now know a great deal about how the cell's living machinery works to make proteins. In addition, they are learning the molecular details of many of the biochemical processes critical to the life of the cell.

Scientists are also making progress in understanding how molecules outside the cell influence what goes on inside and how cells within an organism communicate with each other. One clinical application of this work will probably be better ways to treat chronic wounds and burn injuries.

We now have a much more complete understanding of the molecular signals that cause cells in an embryo to differentiate into muscle, blood, nerve, and other specialized cells. Additional knowledge about cell differentiation will provide greater understanding of normal and defective development in humans.

In recent years, many genes involved in hereditary diseases have been identified. The ability to isolate and copy these genes allows biologists to study what goes wrong in cells to cause the diseases. In addition, researchers are working to determine the sequence of all of the DNA—the genomes—of entire organisms, including humans. This will permit many new studies and should lead to important information on cellular processes and how they are coordinated.

Scientists are working on many techniques to correct faulty genes, including ways to sneak new nucleotide sequences past the body's defense mechanisms. The goal, once the sequences are taken up by the cell, is to get them integrated in such a way that the desired substances are properly made. Gene therapy is also beginning to be employed in new and creative experiments that may someday lead to new ways of treating many different disorders. In the future, it may be possible to use it to genetically engineer living cells to make their own "medicines" in response to carefully controlled chemical signals from outside the cell.

New techniques to rapidly screen chemical compounds are now greatly expanding the pool from which possible therapeutic substances can be drawn. The study of molecular structures by x-ray crystallography has yielded detailed understanding of many molecules critical to health, and may eventually yield therapeutic molecules specifically tailored to "fit" the structures and thus alter their chemical activity. In addition, the science of synthetic chemistry has yielded many improved ways to design new therapeutic substances. The success that all these promising achievements will have in the fight against disease depends on continued progress in understanding cellular biology.

These advances are only a beginning, because the cell still holds many mysteries. It sometimes takes years after a new discovery is made for the potential applications to become clear. Thus, just as no one can predict what basic researchers will discover in the future, neither can the eventual clinical applications of today's results be known.

Scientists now have an unprecedented array of tools and body of knowledge with which to work. A wealth of exciting avenues for scientific exploration are opening. If the momentum can be sustained, the next 50 years may well see victories over many human diseases. ■

Amino Acid A building block of proteins. There are 20 different kinds of amino acids; a protein consists of a specific sequence of amino acids.

Angstrom A unit of length, one hundred-millionth of a centimeter (approximately 0.000000004 inch); used for describing atomic dimensions.

ATP *(adenosine triphosphate)* The compound that serves as a source of energy for the physiological reactions in cells.

Bacterium A one-celled microorganism that contains no nucleus.

Base The basic subunit of DNA or RNA. Paired bases—adenine with thymine and guanine with cytosine (uracil replaces thymine in RNA)—make up each "rung" of the "ladder" of the DNA molecule. See *nucleotide*.

Basic Research Scientific research that seeks to discover how systems work and develop a base of knowledge that other scientists can use in order to achieve practical goals, such as treatments or cures for diseases.

Biochemistry The study of the chemical reactions that occur in living organisms.

Cell The basic subunit of any living organism; the simplest unit that can exist as an independent living system.

Cell Cycle The sequence of events by which the cell duplicates its contents and divides into two.

Cell Surface Membrane A complex film of lipids interspersed with proteins. It covers the cell, maintains its integrity, and controls what goes in and what comes out.

Centrifuge A machine that separates particles according to their size and density by spinning them at varying speeds.

Chloroplast The chlorophyll-containing organelle in green plants in which light energy is converted into sugars.

Cholesterol A waxy lipid produced by animal cells that is a prominent component of cell membranes.

Chromosome A rod-shaped structure containing genes that is found in the cell nucleus. It is composed of DNA and proteins, and can be seen in a light microscope during some stages of cell division.

Codon A sequence of three consecutive nucleotides in a DNA or RNA molecule that codes for 1 of the 20 amino acids in proteins or for a signal to start or stop protein production.

Column Chromatography A technique used to separate the components of biologically active molecules, which move at different speeds through a hollow column that is filled with a chemically reactive material.

Cristae The inward folds of a mitochondrion's inner membrane.

Cyanobacteria *(formerly called blue-green algae)* Single-celled organisms that perform a type of photosynthesis.

Cytoplasm All the substance inside a cell, excluding the nucleus but including the other organelles.

Cytoskeleton A group of non-membrane-bound organelles that supports the cell. Some serve as conduits for the transport of various cell components.

Differentiation The series of biochemical and structural changes that groups of cells undergo in order to form specialized cells and tissues.

DNA *(deoxyribonucleic acid)* The substance of heredity; a large molecule that carries the genetic information necessary for all cellular functions, including the building of proteins. DNA is composed of the sugar deoxyribose, phosphate, and the bases adenine, thymine, guanine, and cytosine.

Electron Microscope A powerful microscope that uses beams of fast-moving electrons instead of light waves to enable objects to be observed.

Endoplasmic Reticulum An organelle made up of membranes that form a system of tubes and flattened sacs. Some of the membranes are smooth (the smooth endoplasmic reticulum); others are rough (the rough endoplasmic reticulum) because they are dotted with ribosomes.

Enzyme A substance (usually a protein) that speeds up, or catalyzes, a chemical reaction without being permanently altered or consumed.

Eukaryotic Cell A cell that has a true nucleus surrounded by a membrane. This group includes all animal and plant cells, except cyanobacteria.

Fluid-Mosaic Model A model of the cell surface membrane in which proteins move about within a bed of semi-fluid lipids.

G Protein One of a group of proteins involved in signal transduction within the cell.

Gel Electrophoresis A technique used to separate molecules according to their sizes and charges.

Gene A unit of heredity; a segment of the DNA molecule containing the code for a specific protein product or function.

Genetic Engineering See *recombinant DNA technology*.

Glycolipid A molecule composed of sugar and fat that forms an important component of cell membranes.

Golgi Apparatus An organelle composed of membranous sacs that packages proteins into vesicles and sends them to the cell's surface or to lysosomes.

Intermediate Filament A component of the cytoskeleton that acts to strengthen the cell.

Ion Any atom or molecule that contains an unequal number of electrons and protons and, therefore, carries a net positive or negative electrical charge.

Ligand Any molecule that binds to a specific site on a protein or other molecule.

Light Microscope An instrument that magnifies objects using curved lenses and white light as a source of illumination.

Lipid A fat or fat-like compound.

Lysosome A small organelle containing powerful enzymes that can digest a variety of materials.

Microfilament A threadlike organelle involved in cell motion, particularly muscle contraction.

Micrometer *(or micron)* One one-thousandth of a millimeter; 10,000 angstroms; convenient for describing the dimensions of cells and organelles.

Microtubule A thin, tubular organelle that acts as a structural support for the cell. During cell division, microtubules form the spindle that directs chromosomes to the daughter cells.

Mitochondrion The cell organelle that converts the energy in sugars into ATP, thereby fueling the cell.

Molecule The smallest physical unit of an element or compound. A molecule of an element consists of one or more identical atoms. A molecule of a compound consists of two or more different atoms.

Multicellular Made up of many cells.

Nanometer One one-thousandth of a micrometer.

Nucleic Acid Either of two kinds of molecules (DNA and RNA), formed by chains of nucleotides, that carry genetic information.

Nucleotide A subunit of DNA or RNA that includes one base, one phosphate molecule, and one sugar molecule (deoxyribose in DNA, ribose in RNA). See *base*.

Nucleus In eukaryotic cells, the membrane-bound organelle that contains the genetic material.

Organelle A specialized structure having a definite function in a cell; for example, the nucleus, a mitochondrion, a ribosome.

Peroxisome A membrane-bound organelle that both generates and breaks down hydrogen peroxide.

Phospholipid A fatty compound that contains phosphate. Phospholipids make up much of the outer membranes of cells and organelles.

Prokaryotic Cell A cell that does not have a membrane around its nuclear region; for example, a bacterium.

Protein A molecule made up of a number of amino acids arranged in a specific order determined by the genetic code. Proteins are essential for all life processes.

Receptor A specialized molecule of a cell's membrane that receives information from the environment and conveys it to other parts of the cell. The information is transmitted in the form of a specific chemical that must fit the receptor like a key in a lock.

Recombinant DNA Technology A body of techniques for cutting apart and splicing together different pieces of DNA. When segments of foreign DNA are transferred into another cell or organism, the substance for which they code may be produced along with substances coded for by the native genetic material of the cell or organism. Thus, these cells become "factories" for the production of the protein coded for by the inserted DNA.

Replication The duplication of hereditary material prior to cell division.

Respiration Within cells, the breakdown of food molecules to liberate metabolically useful energy.

Ribosome An organelle that contains RNA and protein, and is the site of protein synthesis.

RNA (*ribonucleic acid*) A single-stranded nucleic acid that contains the sugar ribose. There are several forms of RNA, including messenger RNA, transfer RNA, and ribosomal RNA (all involved in protein synthesis), as well as several small RNA's whose functions are still being clarified. Certain viruses have RNA, instead of DNA, as their genetic material.

Second Messenger System A multi-step signal amplification process used by the cell to transmit, for example, signals from many hormones that cannot enter the cell directly.

Steroid A molecule related to cholesterol. Many important hormones, such as estrogen and testosterone, are steroids.

Transcription The transfer of information from various parts of the DNA molecule to new strands of messenger RNA, which then carry this information from the nucleus to the cytoplasm.

Translation The conversion of the genetic instructions for a protein from nucleotides of messenger RNA into amino acids.

Vesicle A small, membrane-bound, spherical sac in the cytoplasm of a eukaryotic cell.

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Pages 22, 23 (bottom)—Yunis, J.J., *Human Pathology* 12:494. W.B. Saunders Company, Philadelphia, 1981.

Pages 23, 26, 27 —Watson, J.D., Tooze, J., and Kurtz, D.T., *Recombinant DNA: A Short Course*. W.H. Freeman and Company, New York. 1983.

Pages 29 (top), **30, 32, 37, 46** — Friend, D.S., Brigham and Women's Hospital.

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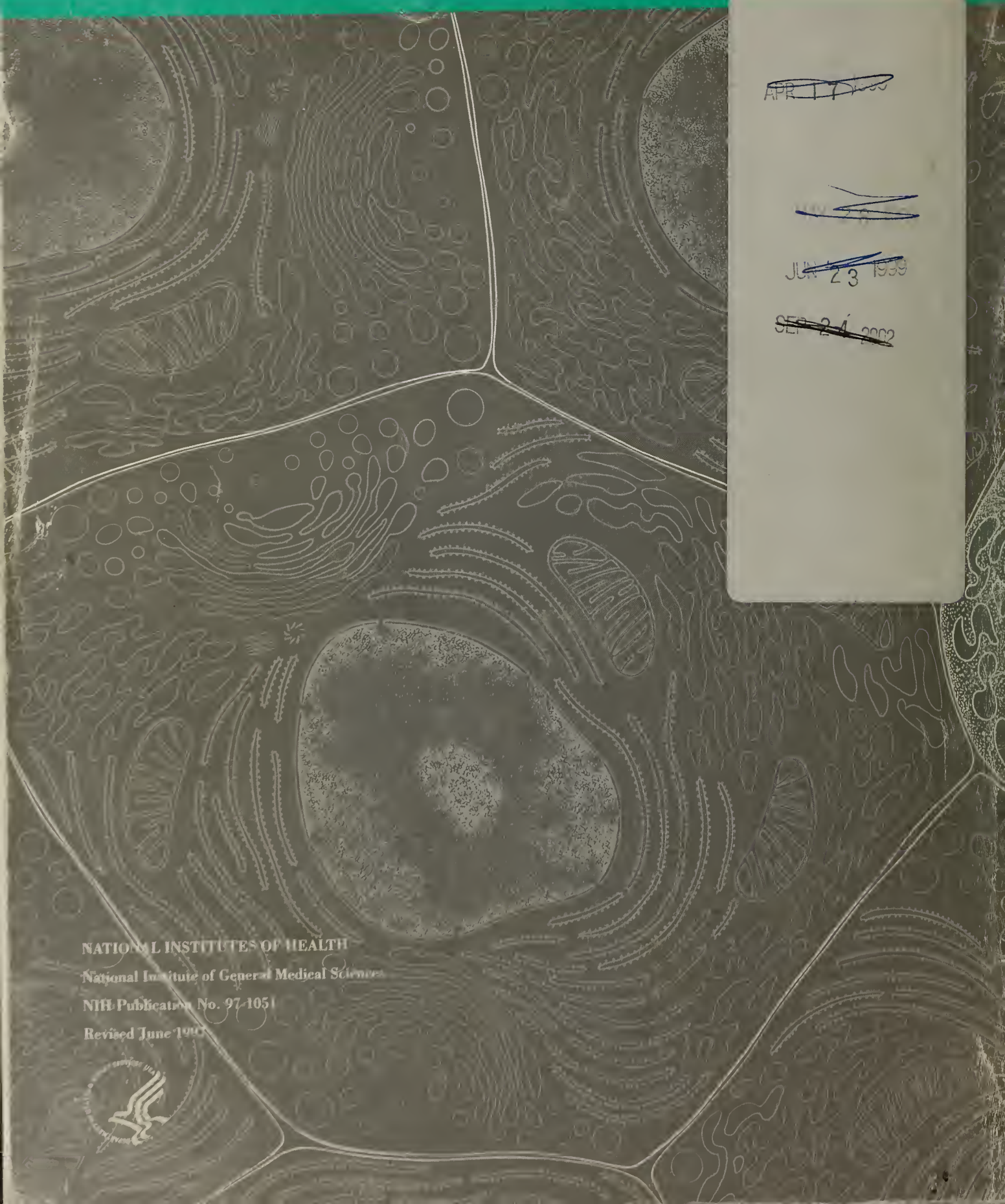

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